

# **STANDARDIZATION AND PHARMACOLOGICAL SCREENING OF *SURANGUSA PARPAM***

The dissertation Submitted by

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Chennai – 47.**

### **DECLARATION BY THE CANDIDATE**

I hereby declare that this dissertation entitled “**Standardization and Pharmacological Screening of *Surangusa Parpam***” is a bonafide and genuine research work carried out by me under the guidance of **Dr.S.Visweswaran M.D(s), Ph.D**, Head of the Department i/c, Department of *Gunapadam*, National Institute of Siddha, Chennai – 47 and the dissertation has not formed the basis for the award of any Degree, Diploma, Fellowship or other similar title.

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### **CERTIFICATE BY THE GUIDE**

This is to certify that the dissertation entitled “**Standardization and Pharmacological Screening of *Surangusa parpam***” is submitted to The Tamilnadu Dr.M.G.R.Medical University, Chennai in partial fulfillment of the requirements for the award of degree of M.D (Siddha) is the bonafide and genuine research work done by **Dr.G.Manikandan** under my supervision and guidance and the dissertation has not formed the basis for the award of any Degree, Diploma, Fellowship or other similar title.

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### **BONAFIDE CERTIFICATE**

This is to certify that the dissertation entitled “**Standardization and Pharmacological Screening of *Surangusa parpam***” is a bonafide work carried out by **Dr.G.Manikandan** a candidate of the National Institute of Siddha, Chennai-47 in partial fulfillment of the University rules and regulations for award of M.D (Siddha) - Gunapadam during the academic year of 2019.

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## 1.INTRODUCTION

The Siddha system dates back to 5000 B.C profounded by Saint Agathiyar and his clan numbering 18 such Siddhars. This system was an amalgam of Tamil literature, culture, tradition, health and many such living forms of 64 types.

"தமிழ்மண் டலமைந்துந் தாவிய ஞானம்  
உமிழ்வது போல வுஅகந் திரிவார்  
அவிழு மனமுமெம் மாதி யறிவுந்  
தமிழ்மண் டலமைந்துந் தத்துவ மாமெ"

- திருமந்திரம்

*Siddha* system of medicine is the oldest in ancient India and was derived by tamil siddhars or spiritual scientists of Tamilnadu. It is a holistic medical science which cares body and mind. Siddhars fundamental principles never differentiated man from the universe. According to them “nature is man and man is nature” and therefore both are essentially one. man is said to be the microcosm and the universe as macrocosm, because what exist in the universe exist in man. Health is an indispensable part of human beings. Siddha traditional system of medicine, we covers and consider expansive possibilities, several diseases and ailments generated from many varying climatic condition and changes.

Siddha system of medicine is a science which treats body and mind. Life style advocacies, selection of functional foods and person oriented treatment regimen are the uniqueness of Siddha system of medicine.

Siddha system of medicine is an integrated part of Indian system, which is a very potent and unique system in existence and practiced in India for thousands of years and above. It is an earliest medical science that stress on positive health, a harmonious blending of physical, mental, social, moral and spiritual welfare of individuals. The Siddha system has developed a rich treasure of medicinal knowledge that includes the use of herbs, metals and minerals, It is a traditional system of medicine which is gradually evolved along with the Dravidian's culture and hence this system is also known as dravidian system of medicine. Siddha system also deals with the concept of salvation in life.

The aim of Siddha medicine to make the body perfect, imperishable and to promote longevity. Siddha system of medicine is considered the oldest documented medicine system of the world. It evolved in south India and the knowledge of siddha medicine was completely flourished in the period of Indus Valley Civilization.

The exponents of Siddha system of medicine are called Siddhars. They are the super human beings with high culture and intellectual abilities. It is considered that Siddha medicine was created by Lord Siva and he is the first Siddhar. There were 18 important Siddhars in olden days and they developed this system of medicine. Siddhars were spiritual adepts who possessed the Ashta siddhis or the eight supernatural powers, They practiced intense yogic practices, including years of periodic fasting and meditation and were believed to have achieved supernatural powers and gained the supreme wisdom and overall immortality.

Through this spiritually attained supreme knowledge, they wrote scriptures on all aspects of life, from arts to science and truth of life to miracle cure for diseases. Food habits and daily activities of an individual play a major role in causing disease. The physical functions of the body is mediated and maintained by three vital forces. They are Vali, Azhal and Iyam. In normal state they are called the forces or Muthathu that sustain and nourish the body. In disease state when the three forces are vitiated they are called Mukkutram. When the three forces are in balance one is healthy.

When vitiated singly or combination bring about disease. Emotion and stress also stimulates the Udal tharhukal (7 physical constitution ending up in a disease. The structural aspect of the human body is said to be "Udal Thathukkal " ( i.e. the physical component of the human body) which consists of seven elements: first is Saaram (Plasma) responsible for growth, development and nourishment; second is Senneer (Blood) responsible for nourishing muscles, imparting colour and improving intellect; the third is Oon (Muscle responsible for shape of the body; fourth is Kollzuppu (Fatty tissue) responsible for oil balance and lubricating joints; fifth is Enbu (Bone) responsible for body structure and posture and movement: sixth is Moolai (Nerve) responsible for strength and the last is Sukilam (Semen) responsible for reproduction.

The functional units of the human body are said to be "Uyir Thathukkal" (i.e. Vatham, Pitham and Kabham). They are considered as three pillars of health and support, the structure and functions of the body. They are involved in regulating all the functions of the body and maintain the balance in the physical, emotional and mental spheres. These Uyir thathukkal co-exist in all the cells of the body. They function in a harmonious manner to create a balance. The factors assumed to affect this equilibrium are environment, climatic conditions, diet, physical activities and stress. The food, which is the basic building material for the human body, gets processed into these body tissues, humors and waste products to determine the balance of the Uyir thathukkal in the body. Siddha medicine means medicine that is perfect.

Siddhars spend their lifetime in experimenting the gifts of Mother Nature the herbs, the minerals and the animals. As a result of their experiments, they could formulate so many valuable medicines which include small herbal preparations to the potent medicines. Herbo-mineral formulation has the metals and minerals uses for chronic disorders in various combinations, dosage forms and at various levels of purities. The traditional medicine is widely used for various human ailments. The usage of herbal medicine could be even traced right from the beginning of mankind. Traditional system of medicines has become significantly more popular all over the globe because of the effective and curative nature for chronic diseases with less toxicity.

Growing population, urbanisation, industrialization, deforestation and increasing number of vehicles results in air pollution, which leads to respiratory diseases. In the past 2-3 decades, respiratory diseases are increased remarkably due to severe environmental pollution. In India, a large number of people are affected by respiratory illness, especially with chronic obstructive type of airway disorder<sup>(43)</sup>. Chronic obstructive lung disease (COPD, Tamil: Swasakasam) is a respiratory disease characterized by cough with expectoration, breath sound like hissing of snake, throat irritation, indigestion, flatulence, redness of the nose, low pitched voice and excessive salivation. Major symptoms are cough, sputum production and exertional dyspnoea frequently of long duration. It is a disease state characterized by air flow limitation that is not fully reversible.

Different conditions of COPD are emphysema (characterized by destruction and enlargement of the lung alveoli), chronic bronchitis (chronic cough and phlegm) and small airway disease (small bronchioles are narrowed). Global initiative for chronic lung disease estimates that COPD will be increased from the sixth most common cause of death worldwide by 2020<sup>(44)</sup>. According to American Centre for Disease control and Prevention (2014), more than 70% of COPD related healthcare expenditure goes to medication and in the US about \$10 billion is being spent annually for hospital care of COPD<sup>(43)</sup>.

In siddha system of medicine many formulations are mentioned for respiratory illness. *Surangusa parpam* is one of the siddha drug mentioned in the siddha text, *Anuboga vaithiya Navaneetham part III pg.no.90*, is useful to treat kapha diseases like kasam (cough, asthma), Suram (fever), and Ulaimanthai (intestinal TB) etc. The ingredients of surangusa parpam are Manosilai (arsenic di sulphide), Sangu (conch), Milagu (pepper).

When traditional literatures were reviewed, it revealed that Manosilai has Anti-pyretic and Anti histaminic properties, Sangu has Anti-inflammatory and Anti pyretic properties and the research articles revealed that the individual ingredients of Surangusa parpam possess Anti-histamine, Anti-inflammatory and Anti pyretic activity but as a finished product no pharmacological activities has been carried out for this formulation. Hence the researcher selected the drug Surangusa parpam to standardize and evaluate the pharmacological activities such as Anti- histamine activity, Anti- inflammatory activity and Anti pyretic activity.

## 2. AIM AND OBJECTIVES

### AIM :

To evaluate the *Standardization and Pharmacological screening of the test drug "Surangusa Parpam"* in an animal model .

### OBJECTIVES :

The following methodology was adopted to standardize and evaluate the pharmacological activities of the test drug-

- Review of various information (siddha and modern) relevant to the study.
- Preparation of the drug as per classical siddha literature.
- Analytical study of the prepared drug
  - Biochemical analysis for determining acidic and basic radicals.
  - Physicochemical analysis
  - Scanning electron microscopy with EDAX
  - UV analysis
  - FT-IR (Fourier Transform Infra-Red) .

### Evaluation of pharmacological activities in animal models

Anti inflammatory activity	-	Carrageenan induced paw oedema method
Antipyretic Activity	-	Brewer yeast induced method
Anti histamine activity	-	Evan's dye method

### 3. MATERIALS AND METHODS:

#### DRUG SELECTION

The drug Surangusa parpam is a siddha formulation mentioned in siddha text Anuboga vaithiya Navaneetham part - III indicated for Suram (fever), Kaasam (cough), Ulaimaanthai (tuberculosis of lung or incurable internal abcess) .

#### INGREDIENTS:

- |   |   |                  |
|---|---|------------------|
| 1. Purified Manosilai (arsenic di sulphide) | - | 4 varagan(14 gm) |
| 2. Purified Milagu (pepper)                 | - | 4 varagan(14 gm) |
| 3. Purified Sangu (conch)                   | - | 4 varagan(14 gm) |

#### Procurement of Raw Drugs:

The raw drugs were procured from a well reputed country shop in Parrys corner, Chennai. All the ingredients were purified and the medicine was prepared in the *Gunapadam* laboratory of National Institute of Siddha.

#### Identification and Authentication of the drug:

- The plant materials were identified and authenticated by the Botanist, Department of Medicinal Botany, National Institute of Siddha.
- The raw drug was authenticated by the Faculty member, Department of Gunapadam, National institute of siddha.

#### Purification of the drugs:

- All the drugs mentioned here were purified as per the Siddha literature.

**METHOD OF PURIFICATION:****PURIFICATION OF MANOSILAI:<sup>(2)</sup>**

Red orpiment (35 grams) was made into small pieces and kept soaked in 175 gms of fermented butter milk in a clay vessel. It was isolated and kindling frequently. In the evening it is washed in the water. The same procedure was repeated for three times to get purified form.

**PURIFICATION OF SANGU:<sup>(2)</sup>**

Take equal quantities of limestone and fullers earth and add eight times of water, put the conch into it and boil well to get it purified.

**PURIFICATION OF PEPPER:<sup>(13)</sup>**

Piper nigrum was soaked in butter milk for 1 ½ hours and then it is dried and roasted to get it purified.

**METHOD OF PREPARATION:**

The above ingredients are soaked in goats urine(2 ½ palam) and kept for 3 days. On fourth day the contents are rubbed for 3 days with the same urine in which they are kept soaked .Then they are made into pellets and dried. The dried pellets are placed in a mud plate which is then covered by a similar mud plate. The margins are covered by a mud pasted cloth, dried and then subjected to pudam with cow dung cakes which are 20 times the weight of sealed mud plates. Again the process is repeated once. Being cooled , the lid is opened and the processed medicine thus obtained is collected and kept in an air tight container.

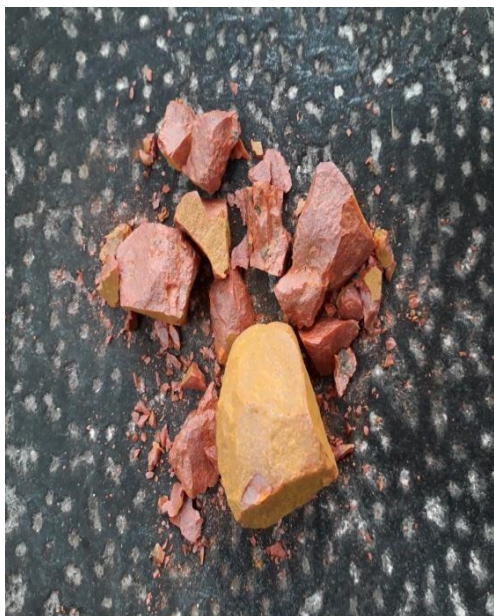
**LABELLING :**

<b>Date of preparation</b>	-	23.04.2018
<b>Name of the preparation</b>	-	Surangusa parpam
<b>Dose</b>	-	One to two kundri(130mg- 260mg), Twice a day, After food
<b>Adjuvant/Vehicle</b>	-	Honey
<b>Route of administration</b>	-	Oral
<b>Duration</b>	-	48days
<b>Indications</b>	-	Suram (fever), kaasam (cough), Ulaimaanthai (tuberculosis of lung or incurable internal abcess)
<b>Date of expiry:</b>	-	100 years
<b>Reference</b>	-	Anuboga vaithiya navaneetham – III pg no 90.



**INGREDIENTS OF SURANGUSA PAMPAM**  
**MANOSILAI**

**BEFORE PURIFICATION**



**AFTER PURIFICATION**



**GOAT URINE**



## PEPPER

**BEFORE PURIFICATION**



**AFTER PURIFICATION**



**SANGU**  
**BEFORE PURIFICATION**



**AFTER PURIFICATION**



## **SURANGUSA PARPAM**



## 4. LITERATURE REVIEW:

### 4.1. GUNAPADAM REVIEW

#### SANGU

<b>Zoological name</b>	:	Turbinella Pyrum
<b>English name</b>	:	sacred chank, conch shell
<b>Synonyms</b>	:	Nandhu, Naagu, Vandu, Sangam, Kodu, Varanam, Idampuri, Valai, Vellai, Devathatham, Kambu, Suthi.

#### General characters:

"கசிவா மிரத்த பித்தங் கண்ணோய்க லேகும்  
பசியாறும் வாதம் பறக்கு - மிசிவுடனே  
தங்கு முளைவிரணந் தாளகலு மேவெள்ளைச்  
சங்கமது வண்டாயிற்றான்"

Sangu is useful in the treatment of Athikuruthi azhutham (hypertension), Kan noi (eye diseases), Valippu (epilepsy) and derangement of vatham humor. It stimulates appetite.

#### MEDICINAL USES:

- ❖ Sangu parpam when given with plant juice of Tabernaemontana divaricate (nanthiyavattam) cures Gunmam (ulcer), Paandu (anaemia), Peru vayiru (ascites), Mega noigal (venereal diseases), Moolam (piles), Maneeral veekam (enlargement of spleen) and Elaippu noi (tuberculosis).
- ❖ Sangu parpam when given with honey cures piles, anaemia, mania and excess saliva.
- ❖ Sangu parpam prepared by daemia extense(uthamani) is used to treat cough, piles, enlarged tonsils, stomach disease, Gunmam, vayu and chest pain.
- ❖ Siddhar theraiyar also reiterates that tuberculosis and kapha disease are cured with conch shell.
- ❖ There is also a practice to prescribe sangu parpam with crunched snail in a conch for curing tuberculosis.
- ❖ Sangu chenduram when given with lemon juice cures white leprosy.

- ❖ Sangu chenduram when given with jiggery it treats ulcer associated with diabetes.
- ❖ The conch is rubbed with breast milk or with *Murraya Koenigii* (Kariveppilai) and applied over the pimples, acne and boils of the eye.
- ❖ Sangu is one of the major ingredient in Vellai mathirai which is used to treat eye related problems.

### **Siddha Formulations Using Sangu As Ingredient:**

#### **1.Sangu parpam :**

Dose : 130mg  
 Adjuvant : Ghee, Butter  
 Indications : Vadha Gunmam, Neer Surukku

#### **1. Sangu chenduram :**

Adjuvant :  
 Sandhana Kuzhambu - Vadham  
 Ulli Rasam - Pitha Noi  
 Vellam - Gunmam

#### **2. Panchakkini chenduram :**

Dose : 3 to 4 Kundri  
 Adjuvant : Ginger Juice, Chukka Kudineer, Milagu Kudineer,  
 Pudhina Kudineer, Neermulli Kudineer.  
 Indications : Gunmam, Indigestion, Kazhichal, Vaayu, Neer Kattu,

#### **3. Naaga Sangu Parpam :**

Indications : Powthiram, Vellai, Vettai, Moolam

#### **4. Vellai Mathirai :**

Indications : Ratha Padalam, Kan Noigal



## **MANOSILAI (Arsenic di sulphide)**

Manosilai is of 2 types.

1. Sivandha aridharam
2. Madal aridharam
3. Kuthiraipal padanam

### **Synonyms:**

Naanmugan, Devi, Naamagal, Bharathi, Vaani, Kalaimagal, Poomagal, Silai, Vil, Sarajothi, Vellachi, Thamarai Vasini.

### **Actions:**

- i. Febrifuge
- ii. Alterative
- iii. Nutrient

### **General characters:**

"கொடிய குஷ்டம் காய்ச்சல் நடுக்கலஜ கல்லிரைப்  
புச்சிலந்திப்பேசுறம் னோசிலைக்குச் பேசு"

(Gunapadam thaathu jeeva vaguppu)

It cures Saruma noi (skin diseases), Suram (fever), Silanthi vidam (spider poisons), Eraippu noi (bronchial asthma), Breathing difficulty, Elaippu noi (Tuberculosis), Kan noi (Eye diseases), Neer erichal (Burning micturition).

"குட்டங்கொடி யரணங்குறுந் திமிர்படையும்  
வட்டமிடுங்கிரந்தி வன்மையெல்லாம் - விட்டுப்போம்  
காய்ச்சல் நடுக்கலஜ கன்னியிரைப் புச்சிலந்தி  
பேச்சுறு மனோசிலையைப் பேண்"

It cures Saruma noi (Skin diseases), Gunmam (Ulcers), Suram (Fever), Nalir (Rigor), Eraippu noi (Bronchial asthma), Silanthi vidam (Spider poison).

## **SIDDHA FORMULATIONS USING MANOSILAI AS INGREDIENT:**

### **1. Vishnu chakara mathirai:**

Dose : 130mg  
Adjuvant : Ginger Juice, Honey, Thirikadugu Chooranam  
Indications : Hemiplegia, Hiccough, Dropsy, Vaayu, Beltching.

### **2. Visha mezhugu**

Dose : 1/4 – 1/2 Kundri  
Adjuvant : Vetrilai Vizhudhu, Sukku Arautha Vizhudhu  
Indications : All Types Of Snake Poison, Rat Poison.

### **3. Kasthuri karuppu**

Dose : 1/2 – 1 Kundri  
Adjuvant : Honey, Mulai Paal, Ginger Juice  
Indications : Cold, Cough, Bronchial Asthma, Fever

### **4. Shaya Gulandhaga chenduram**

Adjuvant : Honey, Thirikadugu Chooranam  
Indications : Kaasam, Shayam, Swasam

### **5. Kaala mega naarayana chenduram**

Dose : 65mg  
Adjuvant : Thippili Powder, Thrikadi Alavu – 15 Days  
Indications : Viranam, Bite Poisons, Cancer, Sinusitis, Ear Diseases, Cardiac Problems.

### **6. Nava Bashana Thailam**

Usage : External Application.  
Indications : All Types Of Vadha Diseases.

### **7. Kabanivarana mathirai**

Indications : Kabakattu

### **8. Putru pathangam**

Dose : 1/4 – 1/2 Kundri  
Adjuvant : Cow Milk  
Indications : Kanna Putru, Maarbu Putru, Kaal Putru, Leucoderma, Oral Cancer.



**Manosilai serum pira marundhugal:**

- Santhirodhaya kuzhambu
- Nayana roga mathirai
- Lekanchana podi
- Thirivanga chenduram
- Rasaveera naga chenduram
- Aanandha bairava mathirai
- Megarajanga mathirai
- Navamoola kuligai
- Thaaba sura mathirai
- Raja boobathi mathirai
- Mantha kaasa mathirai
- Manosilai chooranam
- Kandhaga sudar thailam
- Manosilai ennai
- Visha kuzhambu

**TRADITIONAL USES:**

- ✓ Manosilai is mixed with naayuruvi juice and applied externally for leucoderma.
- ✓ Take 1 varagan of Manosilai, thaalagam, senkottai individually and make into powder form, put into gingelly oil and mixed with white goat urine and boiled well. It is used for pus collected in the ear.
- ✓ It is mixed with some other medicinal powder and used externally for fistula.
- ✓ Manosilai chooranam mixed with siruthekkku and chukku powder and used for bronchial asthma.

**ARSENIC DI SULPHIDE**

Arsenic disulphide is a naturally occurring form of arsenic and is found as realgar, one of the major arsenic containing mineral, arsenic disulphide is insoluble in water and poorly absorbed. It therefore represents a much less acute toxic hazard than soluble arsenic compounds.

**Synonyms:**

Red arsenic sulphide, Arsenic sulphide, Arsenic disulphide, Red orpiment, Ruby arsenic, Realgar.

**Chemical Name and Formula:**

Name : Arsenic di sulphide

Molecular formula :  $\text{As}_2\text{S}_4$

**Physical properties:**

Colour : Red – brown

Melting point :  $320^\circ\text{C}$

Boiling point :  $565^\circ\text{C}$

Solubility : insoluble in water Molecular weight : 213.97

**Preparation:**

It is artificially prepared by fusing arsenious acid 5 parts and sulphur 3 parts.

**Uses:**

- ✓ Used in leather industry, depilatory agent, paint pigment, shoe manufacture, pyrotechnics, rodenticide, (factsheet for realgar) colouring agent in fireworks.
- ✓  $\text{As}_2\text{S}_3$  and  $\text{As}_2\text{S}_4$  have been investigated as treatment for acute promyelocytic leukemia (APL)

**Traditional uses:**

- ✓ Realgar, orpiment, and arsenopyrite provide nearly all the world supply of arsenic as a byproduct of smelting concentrates derived from these ores.
- ✓ Realgar is poisonous. The ancient Greeks, who called it “sandarach”, knew that it was poisonous. It was used to poison rats in medieval Spain and in 16<sup>th</sup> century England (in French)
- ✓ It is still sometimes used to kill weeds, insects and rodents even though more effective arsenic based agents are available.
- ✓ It was along with orpiment, a significant item of trade in the ancient Roman empire was used as a red paint pigment (boston) and a medicine.
- ✓ Other traditional uses include manufacturing shot, printing and dying calico.

## **CHEMICAL ASPECT**

### **PHYSICO CHEMICAL PROPERTIES :**

Chemical structure	-	As <sub>2</sub> S <sub>3</sub>
Molecular Weight	-	213.97
Physical state at room temperature	-	Solid
Colour	-	Red- brown
Odour	-	none
Viscosity	-	NA
PH	-	NA
Solubility	-	Practically insoluble in water
Ignition temperature	-	NIF
Chemical interactions	-	NIF
Major products of combustion	-	Sulphur dioxide gas and Arsenic trioxide
Explosive limits	-	NA
Flammability	-	Ignites at high temperatures
Boiling point	-	565° C
Density	-	Alpha 3.506
Beta	-	3.254
Vapor pressure	-	NIF
Relative vapor density	-	NIF
Flash point	-	NIF
Reactivity	-	No reaction with water

### **MEDICINAL USES:**

- It is purified by being rubbed with the juice of lemons or ginger.
- It is used as an alternative, febrifuge and tonic, given in fever, cough, asthma and skin disease; in these last is used also externally.
- Locally it is applied to fistulous sores recommends for application to the eye, in affections of the internal tunics, tumors or other growths, night blindness etc.,
- It is used as an alternative, febrifuge and tonic, given in fever, cough, asthma and skin disease, in these last is used also externally.
- In fever it is generally used in combination with mercury, orpiment etc..as in following

- Chandesvara Rasa already mentioned under arsenious acid is recommended in Rasenrdrasarasangraha for remittent fevers.
- Svasakuthara Rasa is another preparation mentioned in the same, and consisting of Realgar, Mercury, sulphur, Aconite, Borax, Black pepper, Ginger And Long pepper, is recommended in asthma with cough and in remittent fever with cerebral complications.
- Dose is 4 grains in pills form.
- In coma from remittent fever, these pills are powdered and used as a snuff to rouse the patient.
- A preparation known as chandraprabha varti is made of realgar, gale, conch shell lime, seeds of Maringa pterygosperma, long pepper, liquorices and the kernel of belleric myrobalan in equal parts rubbed together with goat's milk, dried and made in to small pastilles, these are rubbed with a little honey and applied the eyes as a collyrium.
- It is purified by being rubbed with the juice of lime or ginger.
- It is used internally in fever, skin disease, cough ,asthma etc and externally in skin disease realgar mixed with ashes of (achyranthus aspera) is used externally for leucoderma.

#### **MISCELLANEOUS USES:**

- Leather industry
- Depilatory agent
- Paint pigment,
- Shot manufacture,
- Pyrotechnics,
- Rodenticide

## **MILAGU (PEPPER)**

### **Synonyms:**

- Kalinai
- Kari
- Kaayam
- Kolagam
- Thirangal
- Miriyal
- Sarumabantham
- Vallisam
- Maasam
- Kurumilagu
- Malaiyali

**Parts used:** seed, stem, leaves

### **Organoleptic characters:**

Taste : Kaippu, kaarppu

Character : Heat

Division : Kaarppu

### **Actions:**

- Carminative
- Acrid
- Anti periodic
- Rubefacient
- Stimulant
- Resolvent

### **General characters:**

“சீதகூரம் பாண்டு சிலேத்மங் கிராணிகுன்மம்  
வாதம் அருசிபித்தம் மாமுலம் - ஒதுசன்னி  
யாசமபஷ் மாரம் அடன்மேகம் காசமிவை  
நாசங் கறிமிளகினால்”

It cures Nalir suram ( rigor with fever), Kazhichal (diarrhoea), Gunmam (ulcer), Suvaiinmai (aguesia), Paandu (anemia), cold.

**Purification:**

- It is soaked in amla juice to get it purified.
- Piper nigrum is soaked in butter milk for 1 ½ hours and then it is dried and roasted to get it purified.

**Siddha Formulations Using Milagu(pepper) As Ingredient:**

1. **Thrithoda Mathirai**  
Dose : Milagalavu  
Adjuvant : Honey  
Indications : Fever
2. **Swasakudori chooranam**
3. **Kandankathiri chooranam**
4. **Paranjothi kuzhambu**
5. **Magasinthamani kuzhambu**
6. **Paranjothi mathirai**
7. **Maga paranjothi mai**
8. **Malaikkaathan kuligai**
9. **Vida mai**
10. **Sanjaavi kuligai**
11. **Sudu kaadu meetan kuzhambu**
12. **Sinthamani kuligai**
13. **Sangam ver thailam**

## GOAT URINE

### Synonyms:

- Amuri
- Neer
- Moothiram

### Organoleptic characters:

Taste : Thuvarppu, Inippu

### General characteristics:

“வெள்ளாட்டு நீர்க்குப் பேதி மிகவுண்டாங் கிருமி வீக்கம்  
கள்ள மில்லாமற் நீருங் காசில்பா னோய்க ளுக்காம்  
உள்ளுறுகிரந்தி வாத முறுசூலை மந்தம் வாயு  
விள்ளரும் பழைய காய்ச்சல் மேவிய குணமும் போமே”

It cures vatha diseases, Seriyamai (indigestion), Saruma noigal (skin diseases), Soolai, Vaayu, Anemia, Bleeding, Peruvayiru (Ascites) and Suram (Fever).

### Medicinal uses:

- ✓ It is used as Pathiyam.
- ✓ It relieves Dhosas.
- ✓ It cures Bronchial asthma.
- ✓ It relieves pricking pain in the ear.

## **HONEY**

### **Synonyms:**

- Segappu mathu
- Maanira maashidham
- Samberasam
- Amasanam
- Kabilangam
- Minu thavarnam
- Oonin urukinam
- Varna thuppi
- Thooni thailam

### **Organoleptic characters:**

**Taste :** Sweet

### **Actions:**

- Demulcent
- Laxative
- Astringent
- Antiseptic
- Stomachic

### **General characteristics:**

“அனுபான மாப்ப்பின் அவிழ்தமுமாயத் தோன்றி  
கனமான தேகநிலை காட்டிப் பினுமே  
யரசன் முதல்வோ ரையிமாட்டு விதத்தாலே  
பிரசத் தினாற்போம் பிணி”



**Medicinal uses:**

- ✓ It act as a diuretic in children.
- ✓ It is used for making Legium, Mezhugu, Kattu, Kanmai.
- ✓ It is used as a adjuvant for Parpam, Chenduram, Chooranam, Mathirai.
- ✓ It is mixed with lemon juice and used for childrens cough.
- ✓ It is mixed with barley juice and drink, it relieves indigestion, sinusitis, constipation.
- ✓ It act as a cardio tonic for aged persons.
- ✓ It is applied externally for burns.

## 4.2 BOTANICAL REVIEW

### PEPPER :

Pepper is an effective medicinal herb for all the systems of our body, also used as an anti-dote

#### According to Bentham and Hooker classification

Kingdom	-	plantae
Division	-	Angiosperm
Class	-	Dicotyledonae
Order	-	Piperales
Family	-	Piperaceae
Genus	-	Piper
Species	-	nigrum

#### Vernacular names:

Tamil	-	Milagu
Sans	-	Marichan, Ushana, Napusa
Eng	-	Black pepper
Hindi	-	Galumirch
Malayalam	-	Kuru milaku
Telugu	-	Miriyal
Guj	-	kalamari, kalomirchi
Kan	-	kare menasu

#### Description of the plant:

A stout, glabrous, long climber; stem erect, sparingly rooting, thickened at the nodes. Leaves coriaceous, 10-18 cm long, broadly ovate. Acuminate, glabrous. Flowers in slightly interrupted glabrous spikes of variable length 5-15 cm, fruit globose, 6 mm diameter or less, red in colour when ripe.

- Externally it is valued for its Rubefacient properties
- It is used as a local applicant for sore throat, piles and some skin diseases.
- Pepper is much employed as an Aromatic stimulant, cholera, weakness following fever, vertigo, coma.
- In china, pepper is considered as energetic stimulant and carminative.

- The active chemical (piperine) in pepper has been shown to dramatically increase absorption of selenium, vitamin B and b carotene as well as other nutrients.
- Pepper helps to stimulate the secretion of digestive juices known as hydrochloric acid. This acid is responsible for breaking down protein in stomach. If we don't make enough Hcl , we develop conditions like poor digestion, heart burn or symptom of indigestion.
- Pepper has the ability to diminish the amount of gas in the intestinal tract. this leads to less Flatulence and Bloating.
- Pepper contains a good amount of Anti oxidant properties
- Pepper can prevent bacterial growth, especially in the intestinal tract.
- Pepper helps to enhance oxygen supply to the brain. In addition, it may helps to keep the joints and Respiratory system healthy.

#### **DOSING:**

##### **Adults :**

There is no proven effective dose for black pepper in adults. However, nasal inhalation of volatile black pepper oil for one minute up to one month has been studied to help with difficulty in swallowing in stroke patients.

##### **Children**

There is no proven effective dose for black pepper in children.

#### **Chemical constituents<sup>(22)</sup>**

Major constituent of the fruit are alkaloids. They contain upto 9% alkaloids, which include chavicine, b-methyl pyrroline, piperine, piperidine, depentine and piperovatine. It also contain a essential oil with b-bisabolene, caphene, b-caryophyllene and many other terpene and sesquiterpenes, a pungent resin, chavicin, pipertine, lignin, gumpiperyline, piperole A and B. piperanine, monoterpenes, sesquiterpenes, starch and fat. The fruit have also been reported to contain safrole and tannic acid, in addition to other volatile and non volatile constituents.

#### **Part used : Dried unripe fruit**

##### **Action:**

Anti-asthmatic, Resolvent, Antidote, Carminative, Anti-periodic, Rubifacient, Stimulant, Expectorant, Anti-oxidant, Hepato protective

**Medicinal uses:**

- Appetizer- piper nigrum is an effective home remedy for digestive disorders. Mix powdered black pepper with melted palm jiggery increases production of gastric juice and saliva to increase appetite. To get relief from indigestion and stomach heaviness, you can take pepper powder mixed in the butter milk.
- Dentrifice- mix pepper powder and salt to prevent foul breath, dental caries, bleeding and painful gums. Mix pepper powder with clove oil and apply it on the caries to prevent toothache.
- Fever- pepper is effective for cure of fever and severe cold. To get relief from cold, 20 gm of powdered pepper boiled in milk and a pinch of turmeric can be added on it and taken once a day for 3 days.
- Rheumatism- small amount of pepper powder fried in a little amount of sesame oil. This mixture can be applied as an analgesic for the treatment of rheumatic and myalgia pain.
- Impotency-6 pepper are eaten with 4 almonds with milk to treat impotency and it is also a nervine tonic.

### 4.3 ZOOLOGICAL REVIEW :

#### SANGU

<b>Sangu</b>	:	<b>Turbinella pyrum</b>
Zoological name	:	Turbinella pyrua, lam.
Commercial name	:	Sacred chank, conch shell.
Synonyms	:	Xachus pyrua, Gastropoda.

#### Scientific classification

Kingdom	:	Animalia
Phylum	:	mollusca
Class	:	Gastropoda
Order	:	Neogastropoda
Family	:	Turbinellide
Sub family	:	Turbinellinae
Genus	:	Turbinella
Species	:	T.pyrum

#### Vernacular Name

Tamil	:	Sanka
Eng	:	Conch; conch shell
San	:	Shankha
Bengal	:	Sankh

#### Source

Indian ocean coasts

#### External appearance

- A porcelaneous shell is of an oblong or conical form. The oblong form is bulged in the middle and tapering at the end. the conical variety is peculiar. the upper portion is like cork screw, twisted and tapering at the end. The base is broad the interior is hollow.

- The surface is hard of a dull white colour .the upper surface is tuberculate, the under surface is shining, very brittle and translucent highly, chiefly formed of calcium carbonate.
- Male chank measures about 57-60mm in its diameter and female chank about 58-60mm. So usually the chanks measuring minimum 64mm are only picked.

### **Valampuri chank**

Chanks are characterized by large shells with fine texture and are highly valued. Normally, the chank shells are formed in a dextral spiral. Occasionally shells with a sinistral spiral are also formed. This peculiar type of chank is called as “Valampuri chanku

### **ACTION :**

- Expectorant
- Carminative
- Digestive
- Astringent
- Stomachic
- Febrifuge
- Anodyne

**Vamavarta** (“left-turned” as viewed with the aperture uppermost): this is the very commonly occurring dextral form of the species ,where the shell coils or whorls expand in a clockwise spiral when viewed from the apex of the shell.

In Hinduisms, a Dakshinavarta chakra symbolize infinite space and is associated with Vishnu. The vamavarta chakra represents the reversal of the laws of nature and is linked with lord shiva.

### **Actions:**

- Nutrient
- Anodyne
- Carminative
- Stomachic
- Astringent
- Febrifuge
- Expectorant

### Medicinal uses:

- ❖ Diseases of the eye, spasm, kapha diseases, swellings, abdominal pain, distention of abdomen, fever and anaemia are the diseases relieved by chanku.
- ❖ Sangu parpam prepared by daemia extense (uthamani) is used to treat cough, piles, enlarged tonsils, stomach disease, gunmam, vayu and chest pain.
- ❖ Siddhar theraiyar also reiterates that tuberculosis and kapha disease are cured with conch shell.
- ❖ There is also a practice to prescribe sangu parpam with crunched snail in a conch for curing tuberculosis.
- ❖ Sangu chenduram when given with lemon juice cures white leprosy.
- ❖ Sangu chenduram when given with jiggery it treats ulcer associated with diabetes.
- ❖ The conch is rubbed with breast milk or with *Murraya Koenigii* (Kariveppilai) and applied over the pimples, acne and boils of the eye.
- ❖ Sangu is one of the major ingredient in Vellai mathirai which is used to treat eye related problems.
- ❖ Conch shell is used in treating dyspepsia, digestion impairment, malabsorption syndrome hepatomegaly.
- ❖ Wrinkles on skin can be reduced by rubbing with a conch on face and neck after bath, glow of skin will increase naturally.
- ❖ Dark circles under eyes can be cured gently rubbing with conch for 5 minutes per day before sleep.
- ❖ Store some water in a conch overnight and next morning massage your skin with this water. This cures many skin diseases, rashes, allergies etc.
- ❖ Sangu parpam is used for treating diarrhoea (loose stools), acne, pimples, liver enlargement (hepatomegaly), splenomegaly, abdominal pain, indigestion, loss of appetite, heartburn, acid reflux, abdominal distention and irritable bowel syndrome
- ❖ Sangu parpam is used for ear ache, ulcer and eye trouble and is indicated internally in case of dyspepsia, gonorrhea, colic dysentery, jaundice, tympanitis and flatulence.
- ❖ It is given for shooting pain and inflammatory condition in the joints.

**Other uses:**

- Conch shells are sometimes used as decoration ,as decorative, as decorative planters, and in cameo making.
- In classic Mayan art, conches are shown being used in many ways, including as paint and ink holders for elite scribes , as bugles or trumpets, and as hand weapons (held by combatants by inserting 5their hands in the aperture).
- Some American aboriginals used cylindrical conch columella beads as part of breast plates and other personal adornment.

**Test for conch**

- Take well water in a vessel .
- Immerse conch in it for 5 days.
- After 5 days if the colour of the conch remains same it is original .
- If the conch turns into black colour and crack appears on it, that conch is not original.

**GOAT'S URINE**

- Goat's urine is composed mainly of water with little quantities of urea, uric acid, salts (phosphorus, oxalates of sodium, calcium etc) and some hormones in varying proportions. It is although a waste product of the body, none the 3 less it has varying medicinal properties, which had been recognized by the ancient medical practitioners and as such used both internally and externally as medicine.
- From ancient period, apart from cow's urine, in siddha medicine the urine of other animals such as sheep, buffalo, elephant, horse, camel, donkey etc were also very much in use as remedies for the treatment of Worms, Dropsy, Abdominal distention, Flatulence, Colic, Anaemia, Abdominal tumor, Loss of appetite, Tuberculosis, Poison, Haemorrhoids, Amenorrhoea, Leucoderma, Leprosy, Aggravation of kapha and –vatha and in several other mental diseases.



## **URINARY PARAMETERS IN GOAT'S URINE<sup>(26)</sup>:**

<b>COLOUR</b>	Pale yellow, dark brown
<b>URINE VOLUME</b>	10-40ml/kg
<b>SPECIFIC GRAVITY</b>	1.020-1.040
<b>ODOUR</b>	Clear indifferent aromatic
<b>PH</b>	7.5- 8.5
<b>PROTEIN</b>	Negative

## **USES OF GOAT URINE**

- Healing cancer
- Heart disease
- Allergies
- Auto –immune disease
- Diabetes
- Asthma
- Infertility
- Infection and wounds

## **OTHER USES OF GOAT'S URINE<sup>(17)</sup>**

- Goat urine is very useful as an organic fertilizer that is able to establish, maintain soil fertility and crops and can reduce pest attacks,
- It contains known contents like nitrogen(N), potassium(K) and phosphorus (P) in goat urine is higher than from rabbit and cow urine.
- The goat's urine is gently heated and filtered, when it is given at the doses of 1- 1 ½ oz. in *Nardostachys grandiflora* (sodamanjil), it controls epilepsy.

## **GOAT URINE IN AYURVEDA**

- Goat urine is used for its medicinal benefits in Ayurveda. It is used both for oral consumption and external application in itchy skin disorders, Tinea infection etc.
- Urine of the goat is a astringent, sweet, whole some and balances all the three Doshas.

- Goat urine is used as liquid binding agent in Vilwadi Gulika. It is used in treating scorpion bite, rodent bite etc..

#### **Goat urine for external application:**

- Goat urine is applied externally for, itching skin diseases, ringworm, dermatophytosis or tinea infection, Herpes, spreading skin diseases
- Mustard oil cooked with 4 times of goat urine is useful for massage for a patient suffering from epilepsy.

#### **Usage in uterine disorders**

Medicated bougie is prepared of Saussurea lappa, Piper longum, buds of Calotropis gigantea and rock salt by triturating with goat's urine. It is kept inserted into the vagina which cures Karnini type of uterine diseases. All the therapeutic measures prescribed for the treatment of diseases caused by kapha are also beneficial for the cure of this ailment.

#### **HONEY**

Honey is obtained from “bees”, various ingredients of honey helped it to become not only a sweet liquid but also a natural product with high nutritional and medicinal value. the medicinal quality, taste, texture, colour, aroma of honey differs according to the geographical area and species of plants from which it has been collected. Honey is a mixture of sugar and other components.

#### **Typical honey analysis<sup>(24)</sup>**

Fructose	38.5%
Glucose	31%
Sucrose	1%
Water	17%
Maltose	7.2%
Trisaccharides, Carbohydrates	4.2%
Minerals, Vitamins, Enzymes	0.5%

Honey has a density of about 1.36 kgs/liter(36% denser than water)

### Nutritional value per 100 gm

Energy	1272 Kj
Carbohydrate	82.4 g
Dietary fiber	0.2 g
Fat	0 g
Protein	17.10 g
Riboflavin(Vitamin B12)	0.038 mg
Niacin (Vitamin B3)	0.068 mg
Calcium	6 mg
Iron	0.42 mg
Magnesium	2 mg
Phosphorus	4 mg
Sodium	4 mg
Potassium	52 mg
Zinc	0.22 mg

### Medicinal uses of honey<sup>(25)</sup>:

- It blocks the growth of oral bacteria, coats the throat and reduce throat irritation.
- It is effective when used in the treatment of gastric and peptic ulcer.
- It is also considered an Antioxidant. This means that it allows the blood to circulate better and provide more oxygen to areas of the body such as brain.
- It speeds up healing, growth of healing tissue and dries it up.
- Honey has antibacterial properties due to its acidic nature and enzymatically produced hydrogen peroxide.
- Constant use of honey strengthens the white blood corpuscles to fight bacteria and viral diseases. Reduces the effect of poison.

### Precautions to be taken

Honey should not be mixed with hot foods and should not be heated. Honey should not be consumed when working in hot environment, where you are exposed to more heat, should not mixed with rain water. Honey should not be mixed with ghee.

#### **4.4 SCIENTIFIC REVIEW PHARMACOLOGICAL ASPECT**

##### **SANGU:**

##### **Anti-inflammatory activity of sangu parpam<sup>(45)</sup>:**

V. Murugan, et al., was studied that anti inflammatory activity of sangu parpam. Nine healthy albino rats were taken which weighs about 100-150 gm and divided into three groups each consisting of 3 rats. First group was kept as control by giving distilled water of 2ml/100gm body weight. To the second group, the standard drug (Ibuprofen) of 20mg/100gm body weight and to the third group, the test drug Sangu Parpam 1ml/100gm of body weight was given. In this procedure the drug were given daily. Before giving the drug, cotton pellets were prepared of each of 10mg weight and sterilized. Four cotton pellets were kept subcutaneously, in the lower abdomen, two on each side and sutured carefully. After 7 days of drug administration, the animals were anaesthetized. The cotton pellets were found to be surrounded by granulation tissue and were removed and dried. The weight of cotton pellets in each rat was weighed. From this value, the chronic anti-inflammatory action was calculated and compared. This method is more suitable for studying chronic anti-inflammatory action. Sangu parpam possesses 27.8 % chronic Anti-inflammatory effect in rats.

##### **Anti-pyretic study on sangu parpam<sup>(45)</sup> :**

V. Murugan, et al., was studied that anti pyretic activity of sangu parpam. Group of 9 Albino rats were selected and divided equally into three groups. All the rats were made hyperthermic by subcutaneous injection of 12% suspension of yeast at a dose of 1ml/100mg of body weight. 10 hours later, one group of animals were given test drug by gastric tube in a dose of 1.2mg/100gm, the second group received only distilled water in a dose of 1ml and the third group received 30mg/100gm of Sodium Salicylate as the standard. The mean rectal temperature for the three groups was recorded at 0 hr, 1½ hrs, 3 hrs, 4½ hrs after the drug administration. The difference between the mean temperature of the control group, standard and that of the other group is measured. The test drug Sangu Parpam has got significant Anti-pyretic activity on rats.

**Analgesic activity of sangu parpam<sup>(34)</sup>:**

Three groups of rats on either sex were divided into three groups consisting of three rats each. First group was given 2ml of water and kept as control. Second group was administered with pethidine (1 mg/kg bodyweight) intra-peritoneally. The test drug sangu parpam at the dose of 1 ml/100gm body wt (25 mg of sangu parpam was dissolved in 10ml of honey and 10ml of water), was administered to the group. The test drug sangu parpam has got mild analgesic action.

**Anti-microbial activity of sangu parpam<sup>(34)</sup>:**

Sangu parpam has mild inhibitory activity against E.coli, Klebsiella, proteus, pseudomonas and staphylococci in 30 mg/10 ml, 40mg/10 ml and 50 mg/10 ml concentrations.

**Anti-ulcer activity of sangu parpam<sup>(34)</sup>:**

Male wistar rats of weight (175±5gm) were selected. Animals of control group received saline (5 ml/kg) and test groups received sangu parpam (25 mg/kg, 50mg/kg) for 6 days. From day 6, the animals received saline or test drug, 2 hrs prior to the administration of indomethacin (20 mg/kg, orally). Overnight fasted animals were sacrificed by cervical dislocation 3 hrs after the last dose of ulcerogen. The stomach was incised along the greater curvature and examined for ulcer. The response of the parpam on ulcer index, lipid peroxidation (thio barbituric acid reacting substances TBARS) in gastric tissue and serum calcium was determined. Sangu parpam caused significant reduction in ulcer index ( $p < 0.001$ ) in both the indomethacin and cold restraint models.

## **PIPER NIGRUM**

### **Analgesic activity of Piper nigrum<sup>(36)</sup> :**

L. Farhana Tasleem, et al., was studied the analgesic activity of piper nigrum. Piperine at a dose of 5 mg/kg and ethanol extract at a dose of 15 mg/kg after 120 min and hexane extract at a dose of 10 mg/kg after 60 min exhibited significant ( $P < 0.05$ ) analgesic activity by tail immersion method, in comparison to ethanol extract at a dose of 10 mg/kg using analgesy-meter in rats. However, with hotplate method, piperine produced significant ( $P < 0.05$ ) analgesic activity at lower doses (5 and 10 mg/kg) after 120 min. A similar analgesic activity was noted with hexane extract at 15 mg/kg. However, in writhing test, ethanol extract significantly ( $P < 0.05$ ) stopped the number of writhes at a dose of 15 mg/kg, while piperine at a dose of 10 mg/kg completely terminated the writhes in mice. It is concluded from the present study that Piper nigrum L possesses potent analgesic activity.

### **Anti-Inflammatory Activities Of Piper Nigrum<sup>(38)</sup>:**

In the evaluation of anti-inflammatory effect using plethysmometer, piperine at doses of 10 and 15 mg/kg started producing anti-inflammatory effect after 30 min, which lasted till 60 min, whereas hexane and ethanol extracts also produced a similar activity at a slightly low dose (10 mg/kg) but lasted for 120 min. It is concluded from the present study that Piper nigrum L possesses anti-inflammatory activity.

### **Analgesic, Antipyretic and Ulcerogenic Effects of Piperine<sup>(46)</sup>:**

Evan Prince Sabinaa, et al., was studied the Analgesic, Antipyretic and Ulcerogenic activity of Piperine. Mice were administered piperine (20 and 30 mg/kg) intraperitoneally; hot plate reaction test and acetic acid test were used to determine the analgesic activity of piperine in mice. Antipyretic and ulcerogenic effects of piperine were also evaluated. It was found that piperine exhibits significant analgesic and antipyretic activities without ulcerogenic effects. The results were comparable with indomethacin which was used as standard drug for reference. The results thus obtained prove the analgesic effects of piperine.

**MANOSILAI:****Anti -Inflammatory Activity Of Pancha Pashana Chendhulam<sup>(47)</sup>:**

Rajanandhini M\*,et al., was studied the anti inflammatory activity of pancha pashana chendhulam. The anti-inflammatory was done by carrageenan induced hind paw edema method using plethysmometer. Indomethacin used as a standard drug. For this activity test groups received Control, Induced 1% Carrageenan (0.1 ml), Standard Indomethacin (40mg/kg) and the PPCM in 10mg/kg and 20mg/kg. The anti-inflammatory activity is more effective in Group V Carrageenan induction with oral administration of PPCM 20mg/kg (40.08%) compared to Group IV Carrageenan induction with oral administration of PPCM 10mg/kg (23.49%). The standard drug indomethacin showed 44.50% inhibition of paw edema. The results suggested that the PPCM has exhibited an effective anti-inflammatory activity mediated via either by inhibition of cyclooxygenase cascade and by blocking the release of vasoactive substances like histamine, serotonin and kinins.

## **RESEARCH ABOUT URINE:**

### **Antigenotoxic And Anticlastogenic Properties<sup>(26)</sup>:**

Dutta et al. (2006) studied the anticlastogenic effect of redistilled goat urine distillate (RCUD) in human peripheral lymphocytes (HLC) challenged with manganese dioxide and hexavalent chromium. The redistilled cow's urine distillate possesses strong antigenotoxic and anticlastogenic properties against HPNLs and HLC treated with Cr+6 and MnO<sub>2</sub>. This property is mainly due to the antioxidants present in RCUD.

### **Antioxidant Effect Of Goat Urine<sup>(25)</sup>:**

The redistillate of goat urine was found to possess total antioxidant status of around 2.6  $\mu$ mol, contributed mainly by volatile fatty acids (1500 mg/L) as revealed by the GC-MS studies. These fatty acids and other antioxidants might cause the observed protective effects (Krishnamurthi et al., 2004).

### **In Vitro Anti-Microbial Activity Of Goat Urine Peptides<sup>(26)</sup>:**

Vaibhav Tomar, et al., was studied the invitro anti microbial activity of goat urine. The results of RDA revealed significant antimicrobial activity in fraction containing cationic peptides against *S. aureus* and *E. coli* which showed 23 mm and 26 mm of zone of inhibition, respectively. While No antimicrobial activity was observed in all other fractions containing neutral/anionic peptides as evident by absence of zone of inhibition. The result of determination of MIC value of cationic urinary peptides of goats against *E. coli* and *S. aureus* revealed was observed 0.039  $\mu$ g/ $\mu$ l and 0.0199  $\mu$ g/ $\mu$ l, respectively. The results of the RDA clearly indicated that the cationic peptides exhibited significant inhibitory potential against *S. aureus* and *E. coli*.



#### **4.5. PHARMACEUTICAL REVIEW:**

##### **PARPAM:**

**Parpam** is equivalent to calyx, which is prepared by a process of calcination. Parpam is apparently a tamilized form of the Sanskrit word bhasma. Parpam has held the ground in siddha medicine. Parpam is prepared from different sources like metals, minerals, marine products etc...

##### **PURIFICATION OF THE RAW DRUG**

The following processes are involved

- Elimination of harmful matter from the drug.
- Modification of undesirable physical properties of the drug.
- Conversion of some of the characteristics of the drug to different stages.
- Enhancement of the therapeutic action.

##### **Parpam- nano particle**

Animal derivatives such as horns, shells, feathers, metallic and nonmetallic minerals are normally administered as parpam. Parpam means an ash obtained through incineration. The starter material undergoes an elaborate process of purification followed by the reaction phase, which involves incorporation of some other mineral and herbal extracts. Then the material in pellet form is incinerated in a furnace. For the complete transformation of the material into the parpam state, the process of grinding, drying, and calcification may have to be repeated several times or atleast as many times as directed in the recipe. However the calcination is repeated until a satisfactory product is obtained. But in those instances where the number of calcinations is definitely indicated the process should be repeated accordingly, even if a satisfactory parpam is obtained within a few calcifications.

While preparing the parpams of lead, tin, and zinc, the number of dung cakes used as fuel, should always be comparatively lesser than the number used for other metals, because excessive heating will result in the reversion of the parpam to the metallic state.

##### **Physical characters Colour :**

A specific color is mentioned for each parpam. They are generally white and pale. The color of the preparation primarily depends on the parent material.

**Lusterless :**

Parpam must be lusterless before therapeutic application. For this test, parpam is observed under bright sunlight whether luster is present or not, if luster is still present it indicates further incineration.

**Lightness and fineness :**

Parpam floats on stagnant water surface. This test is based on law of surface tension. Properly incinerated parpam need to float on water surface.

**Tactile sensation :**

Tactile sensation can be absorbed and assimilated in the body without producing any irritation to mucous membrane of gastro intestinal tract.

**Particle size :**

Prepared parpam should be in powder form. Particle of parpam should be like pollen grains of pondanus odoratissimus flower. Physiologically, the particle fineness is of great importance. Most compounds of metals and minerals are not absorbed by the body from the digestive tract, because under ordinary circumstances, these substances could not be reacted upon by the secretions of the digestive system, so as to render them absorbable by the organism. This difficulty is overcome when the individual particles of these compounds are very minute. This concurrently has a say in the matter of dosage in that the dose could be reduced to a great degree as a major part of the finely particulate drug is absorbed into the system.

**Quality control of parpam :**

Traditionally, the end points of incineration of a metal and its conversion to a parpam state are evaluated based on the following criteria.

- When a parpam is spread between the index finger and thumb and rubbed, it should be so fine as to get easily into the lines and crevices of the fingers and should not be washed out from the lines of the fingers.
- When a small quantity is spread on cold and still water, it should float on the surface.
- The parpam should not revert to the original state.
- Parpam should be tasteless.
- The parpam should not produce nausea when administration.

- The parpam if satisfactory completed, is irreversible to its metallic waste when heated with a mixture of cane jaggery, hemp powder, ghee and honey.

**Importance of parpam :**

- ❖ Maintain optimum alkalinity for optimum health.
- ❖ Provide easily absorbed and usable calcium.
- ❖ Cleanse the kidneys, intestines and liver.
- ❖ Maintain stronger bones and healthier teeth.
- ❖ Alleviate insomnia, depression.
- ❖ Keeps rhythmic heart beating.
- ❖ Keeps arrhythmias and minerals balance.
- ❖ Help metabolize iron in body.
- ❖ Aid nervous system.
- ❖ Breakdown heavy metals and drug residues in body.
- ❖ Neutralize harmful acids that lead to illness.
- ❖ Achieve a healthy alkaline level by neutralizing acid.
- ❖ Protect body from free radical damage.

**Storage of parpam :**

- ✓ Parpams are usually stored in glass bottles.
- ✓ For smaller packing, vials of glass may be employed.
- ✓ It is highly desirable that these preparations be stored and retained in relevant labelled containers.
- ✓ They are said to retain their potency for 100 years, if properly stored.

## **5. ANALYTICAL STUDIES OF SURANGUSA PARPAM**

Analytical study of the prepared drug brings the validation to be used as a medicine by subjecting the drug to many analysis and determining its quality and effectiveness. Standardization includes many studies such as its organoleptic properties, physico chemical properties and to assess the active principles and elements present in the drug. Thus analytical study brings the efficacy and potency of the drug.

As per AYUSH protocol for analytical study, the following parameters were evaluated.

### **Organoleptic characters:**

- ❖ Colour
- ❖ Odour
- ❖ Taste
- ❖ Texture

### **Physico chemical analysis:**

- ❖ Determination of ash values
- ❖ Physical characterization

### **Chemical analysis:**

Preliminary basic and acidic radical studies.

### **Elemental analysis:**

- ❖ SEM and EDAX analysis
- ❖ FT-IR
- ❖ UV spectroscopy analysis

### **5.1. ORGANOLEPTIC CHARACTERIZATION OF SURANGUSA PARPAM**

The organoleptic characters of the sample drug were evaluated. 1gm of the test drug was taken and the following characters were seen.

Colour, odour, taste, texture and other morphology were viewed by naked eye under sunlight, then the result was noted.

#### **Colour:**

The medicine was taken into watch glasses and placed against white background in white tube light. It was observed for its colour by naked eye.

#### **Odour:**

The medicine was smelled individually. The time interval among two smelling was kept 2 minutes to nullify the effect of previous smelling.

#### **Results:**

The results of organoleptic character were showed in table-1

## 5.2 THE PRELIMINARY PHYSICOCHEMICAL SCREENING TEST OF *SURANGUSA PARPAM*

Physicochemical Properties of *surangusa parpam* was carried out for each extracts of *surangusa parpam* as per the standard procedure at The Tamil Nadu Dr. MGR Medical University, Anna Salai, Guindy, Chennai-600032.

Physico-chemical studies of the plant drugs are necessary for standardization, as it helps in under-standing the significance of physical and chemical properties of the substance being analyzed in terms of their observed activities and especially for the determination of their purity and quality. The analysis includes the determination of ash value, Loss on drying of the sample at 105°C, pH value and Extractive value. These were carried out as per guidelines.

### 1. Loss On Drying:

An accurately weighed 2g of *surangusa parpam* formulation was taken in a tarred glass bottle. The crude drug was heated at 105°C for 6 hours in an oven till a constant weight. Percentage moisture content of the sample was calculated with reference to the shade dried material.

### 2. Determination of total ash:

Weighed accurately 2g of *surangusa parpam* formulation was added in crucible at a temperature 600°C in a muffle furnace till carbon free ash was obtained. It was calculated with reference to the air dried drug.

### Calculation:

$$\text{Percentage of total ash} = \frac{\text{Weight of the ash}}{\text{Weight of test drug taken}} \times 100$$

### 3. Determination of acid insoluble ash:

Ash above obtained, was boiled for 5min with 25ml of 1M Hydrochloric acid and Filtered using an ash less filter paper. Insoluble matter retained on filter paper was washed with hot water and filter paper was burnt to a constant weight in a muffler furnace. The percentage of acid insoluble as was calculated with reference to the air dried drug.

**Calculation:**

$$\text{Percentage of acid-insoluble ash} = \frac{\text{Weight of the acid-insoluble residue}}{\text{Weight of test drug taken}} \times 100$$

**4. Determination of water soluble ash:**

Total ash 1g was boiled for 5min with 25ml water and insoluble matter collected

on an ash less filter paper was washed with hot water and ignited for 15min at a temperature not exceeding 450°C in a muffle furnace. The amount of soluble ash is determined by drying the filtrate.

**5. Determination of water soluble Extractive:**

5gm of air dried drug, coarsely powered *surangusa parpam* was macerated with 100ml of distilled water in a closed flask for twenty-four hours shaking frequently.

Solution was filtered and 25 ml of filtrate was evaporated in a tarred flat bottom shallow dish, further dried at 100°C and weighted. The percentage of water soluble extractive was calculated with reference to the air dried drugs.

**Calculation:**

$$\text{Percentage of water soluble extract} = \frac{\text{Weight of the extract}}{\text{Weight of sample taken}} \times \frac{100}{25}$$

**6. Determination of alcohol soluble extractive:**

2.5gm. of air dried drugs; coarsely powdered *surangusa parpam* was macerated with 50 ml. alcohol in closed flask for 24 hrs. With frequent shaking it was filtered rapidly taking precaution against loss of alcohol. 10ml of filtrate was then evaporated in a tarred flat bottom shallow dish, dried at 100°C and weighted. The percentage of alcohol soluble extractive was calculated with reference to air dried drug.

**Calculation:**

$$\text{Percentage of alcohol soluble extract} = \frac{\text{Weight of the extract}}{\text{Weight of sample taken}} \times \frac{100}{25}$$

**7. Determination of pH:**

Five grams of *surangusa parpam* was weighed accurately and placed in clear 100 ml beaker. Then 50 ml of distilled water was added to it and dissolved well. After 30 minutes it was then applied in to pH meter at standard buffer solution of 4.0, 7.0, and 9.2. Repeated the test four times and average was recorded.

The results were tabulated in Table –02



### 5.3.CHEMICAL ANALYSIS OF SURANGUSA PARPAM

The chemical analysis of Surangusa parpam was carried out in Bio chemistry lab, National Institute of Siddha, Tambaram sanatorium .

S.No	EXPERIMENT	OBSERVATION	INFERENCE
1.	Physical Appearance of extract	White in colour	
2.	<b>Test for Silicate</b> A 500mg of the sample was shaken well with distilled water.	Sparingly soluble	Presence of Silicate
3.	<b>Action of Heat:</b> A 500mg of the sample was taken in a dry test tube and heated gently at first and then strong.	No White fumes evolved.	Absence of Carbonate
4.	<b>Flame Test:</b> A 500mg of the sample was made into a paste with Con. HCl in a watch glass and introduced into non-luminous part of the Bunsen flame.	bluish green flame appears	presence of copper
5.	<b>Ash Test:</b> A filter paper was soaked into a mixture of extract and dil. cobalt nitrate solution and introduced into the Bunsen flame and ignited.	Appearance of yellow color flame	Absence of sodium

#### Preparation of Extract:

5gm of Surangusa parpam was taken in a 250ml clean beaker and added with 50ml of distilled water. Then it was boiled well for about 10 minutes. Then it was cooled and filtered in a 100ml volumetric flask and made up to 100ml with distilled water. This preparation was used for the qualitative analysis of acidic/basic radicals and biochemical constituents in it.

S. No	EXPERIMENT	OBSERVATION	INFERENCE
	<b>I. Test For Acid Radicals</b>		
1.	<b>Test For Sulphate:</b> 2ml of the above prepared extract was taken in a test tube to this added 2ml of 4% dil ammonium oxalate solution	Cloudy appearance	Presence of Sulphate
2.	<b>Test For Chloride:</b> 2ml of the above prepared extract is added with 2ml of dil-Hno <sub>3</sub> until the effervescence ceases off. Then 2ml of silver nitrate solution is added.	Cloudy appearance was formed	Presence of Chloride
3.	<b>Test For Phosphate:</b> 2ml of the extract is treated with 2ml of ammonium molybdate solution and 2ml of Con.HNO <sub>3</sub>	Cloudy yellow appearance present	Presence of Phosphate
4.	<b>Test For Carbonate:</b> 2ml of the extract was treated with 2ml dil. magnesium sulphate solution.	Cloudy appearance was evolved.	presence of carbonate
5.	<b>Test For Nitrate:</b> 1gm of the extract was heated with copper turnings and concentrated H <sub>2</sub> SO <sub>4</sub> and viewed the test tube vertically down.	No Brown gas was evolved	Absence of nitrate
6.	<b>Test For Sulphide:</b> 1gm of the extract was treated with 2ml of Con. HCL	rotten egg smelling gas was evolved	presence of Sulphide
7.	<b>Test For Fluoride &amp; Oxalate:</b> 2ml of extract was added with 2ml of dil. Acetic acid and 2ml dil.calcium chloride solution and heated.	cloudy appearance occurs.	presence of fluoride and oxalate

8.	<b>Test For Nitrite:</b> 3drops of the extract was placed on a filter paper, on that-2 drops of dil.acetic acid and 2 drops of dil.Benzidine solution were placed.	No characteristic changes were noted.	Absence of nitrite
9.	<b>Test For Borate:</b> 2 Pinches (50mg) of the extract was made into paste by using dil.sulphuric acid and alcohol (95%) and introduced into the blue flame.	No Appearance of bluish green color.	Absence of borate
<b>II. Test For Basic Radicals</b>			
1	<b>Test For Lead:</b> 2ml of the extract was added with 2ml of dil.potassium iodine solution.	No Yellow precipitate was obtained	Absence of lead
2	<b>Test For Copper:</b> One pinch (25mg) of extract was made into paste with Con. HCl in a watch glass and introduced into the non-luminous part of the flame.	blue colour appeared	presence of copper
3	<b>Test For Aluminium:</b> To the 2ml of extract dil.sodium hydroxide was added in 5 drops to excess.	yellow Colour appeared characteristic changes chara	presence of Aluminium.
4	<b>Test For Iron:</b> a. To the 2ml of extract, added 2ml of dil.ammonium thiocyanate solution b. To the 2ml of extract 2ml thiocyanate solution and 2ml of con HNO <sub>3</sub> is added	Mild Red colour appeared  Blood red colour appeared	presence of Iron  presence of iron.
5	<b>Test For Zinc:</b> To 2ml of the extract dil. sodium hydroxide solution was added in 5 drops to excess and dil. ammonium chloride was added.	White precipitate was formed	presence of Zinc

6	<b>Test For Calcium:</b> 2ml of the extract was added with 2ml of 4% dil.ammonium oxalate solution	Cloudy appearance and white precipitate was formed	presence of calcium
7.	<b>Test For Magnesium:</b> To 2ml of extract dil. sodium hydroxide solution was added in 5 drops to excess.	White precipitate was obtained	presence of magnesium
8.	<b>Test For Ammonium:</b> To 2ml of extract 1 ml of Nessler's reagent and excess of dil.sodium hydroxide solution are added.	Brown colour appeared	presence of ammonium
9.	<b>Test For Potassium:</b> A pinch (25mg) of extract was treated with 2ml of dil. sodium nitrite solution and then treated with 2ml of dil. cobalt nitrate in 30% dil. glacial acetic acid.	Yellow precipitate was obtained	presence of potassium
10.	<b>Test For Sodium:</b> 2 pinches (50mg) of the extract was made into paste by using HCl and introduced into the blue flame of Bunsen burner.	No yellow colour flame evolved. of yellow colour flame	Absence of sodium
11.	<b>Test For Mercury:</b> 2ml of the extract was treated with 2ml of dil. sodium hydroxide solution.	No Yellow precipitate was obtained	Absence of Mercury
12.	<b>Test For Arsenic:</b> 2ml of the extract was treated with 2ml of dil. sodium hydroxide solution.	No Brownish red precipitate was obtained	Absence of arsenic

	<b>III. Miscellaneous</b>		
1.	<b>Test For Starch:</b> 2ml of extract was treated with weak dil.Iodine solution	Blue colour developed	presence of starch
2.	<b>Test For Reducing Sugar:</b> 5ml of Benedict's qualitative solution was taken in a test tube and allowed to boil for 2 minutes and added 8 to 10 drops of the extract and again boil it for 2 minutes. The colour changes were noted.	No Brick red colour is developed	Absence of reducing sugar
3.	<b>Test For The Alkaloids:</b> a) 2ml of the extract was treated with 2ml of dil.potassium Iodide solution. b) 2ml of the extract was treated with 2ml of dil.picric acid. c) 2ml of the extract was treated with 2ml of dil.phosphotungstic acid.	Yellow colour developed  White precipitate developed	Presence of Alkaloid
4	<b>Test For Tannic Acid:</b> 2ml of extract was treated with 2ml of dil. ferric chloride solution	black precipitate was obtained	presence of Tannic acid
5	<b>Test For Unsaturated Compound:</b> To the 2ml of extract, 2ml of dil. Potassium permanganate solution was added.	Potassium permanganate is not decolourised	Absence of unsaturated compound
6	<b>Test For Amino Acid:</b> 2 drops of the extract was placed on a filter paper and dried well. 20ml of Burette reagent was added.	No Violet colour appeared	Absence of amino acids

7	<b>Test For Type of Compound:</b> 2ml of the extract was treated with 2 ml of dil. ferric chloride solution.	No green and colour developed	Absence of quinolepinephrinepy rocatechoantipyrine
		No Red colour developed	Aliphatic amino acid and meconic acid are absent.
		No Violet colour developed	Apomorphine salicylate and Resorcinol were absent
		No Blue colour developed.	Morphine, Phenol cresol and hydrouinone were Absent.

## **INSTRUMENTAL ANALYSIS**

### **5.4. FT-IR (Fourier Transform Infra-Red)**

The fourier transform infrared spectroscopy test was carried out for surangusa parpam as per the standard procedure. The experimental procedure was done CECRI, karaikudi.

#### **DEFINITION:**

FTIR offers quantitative and qualitative analysis for organic and inorganic samples. Fourier Transform Infrared Spectroscopy (FTIR) identifies chemical bonds in a molecule by producing an infrared absorption spectrum. The spectra produce a profile of the sample, a distinctive molecular fingerprint that can be used to screen and scan samples for many different components. FTIR is an effective analytical instrument for detecting functional groups.

#### **DESCRIPTION:**

The perkin elmer spectrum FTIR instrument consists of globar and mercury vapour lamp as sources, an interferometer chamber comprising of KBr and mylar beam splitters followed by a sample chamber and detector. Entire region of 400-4500  $\text{cm}^{-1}$  is covered by this instrument. The spectrometer works under purged conditions. Solid samples are dispersed in KBr or polyethylene pellets depending on the region of interest. This instrument has a typical resolution of 1.0  $\text{cm}^{-1}$ . signal averaging, signal enhancement, base line correction and other spectral manipulations are possible. The interference pattern obtained from a two beam interferometer as the path difference between the two beams is altered, when fourier transformed, gives rise to the spectrum. The transformation of the interferogram into spectrum is carried out mathematically with a dedicated on line computer.

**APPLICATIONS:** Quantitative Scans, Qualitative Scans x Solids, Liquids, Gases

- ❖ Organic Samples, Inorganic Samples
- ❖ Unknown Identification
- ❖ Impurities Screening
- ❖ Formulation
- ❖ Pharmaceuticals.

**Fig 1.FTIR ( Fourier Transform Infrared Spectroscopy)**



## **FTIR INSTRUMENT**

### **INSTRUMENT DETAILS**

Model : Spectrum one: FT-IR Spectrometer Scan Range : MIR 450-4000  $\text{cm}^{-1}$   
Resolution : 1.0  $\text{cm}^{-1}$   
Sample required : 50 mg, solid or liquid.

### **Sample preparation:**

Solid: KBr or nujol mull method Liquid: cal / TlBr cells

Gas: Gas cells.

### **KBr method:**

The sample was grounded using an agate motor and pestle to give a very fine powder. The finely powder sample was mixed with about 100 mg dried potassium bromide salt. The mixture was then pressed under hydraulic press using a die to yield a transparent disc (measure about 13mm diameter and 0.3 mm in thickness) through which the beam of spectrometer passed.

Infrared spectrum is useful in identifying the functional groups like  $\text{-OH}$ ,  $\text{-CN}$ ,  $\text{-NH}_2$ , etc. also quantitative estimation is possible in certain cases for chemical, pharmaceuticals, petroleum products etc. resins from industries, water and rubber samples can be analysed. Blood and food materials can also be analysed.



**Measurements techniques:**

The procedure for recording the %T or %A is as follows:

1. Air is first scanned for the reference and stored. The sample is then recorded and finally the ratio of the sample and reference data is computed to give required %T or %A at various frequencies.
2. Study of substances with strong absorbance bands and weak absorbance bands as well as possible.
3. Small amount of samples are sufficient. 4. High resolution is obtained.

**Procedure:**

Typically, 1.5 mg of protein, dissolved in the buffer used for its purification, were centrifuged in a 30K centrifuge on micro concentrator (Amicon) at 3000 g at 4°C until a volume of approximately 40  $\mu$ l.

1. Then, 300  $\mu$ l of 20 mM buffer, prepared in H<sub>2</sub>O or 2 H<sub>2</sub>O, pH or pD 7.2, were added and the sample concentrated again. The pD value corresponds to the pH.
2. Meter reading  $\pm 0.4$ . the concentration and dilution procedure was repeated several times in order to completely replace the original buffer with this buffer.
3. The washings took 24h, which is the time of contact of the protein with the 2 H<sub>2</sub>O.
4. Medium prior FT-IR analysis, in the last washing, the protein was concentrated to a volume of approximately 40  $\mu$ l and used for the infrared measurements.
5. The concentrated protein sample was placed in CaF<sub>2</sub> windows and a 6 mm tin spacer or a 25mm Teflon spacer for the experiments in H<sub>2</sub>O or 2 H<sub>2</sub>O, respectively. FT-IR spectra were recorded by means of a Perkin – Elmer – Spectrum – 1 FT-IR spectrometer using a deuterated triglycine sulfate detector.
6. At least 24 h before, and during data acquisition, the spectrometer was continuously purged with dry air at a dew point of 40°C. Spectra of buffers and samples were acquired at 2 cm<sup>-1</sup> resolution under the same scanning and temperature conditions. In the thermal denaturation experiments, the temperature was raised in 5°C steps from 20 to 95°C.
7. Before spectrum acquisition, samples were maintained at the desired temperature for the time necessary for the stabilization of temperature inside the cell (6min). Spectra were collected and processed using the SPECTRUM software from Perkin-Elmer.

Correct subtraction of H<sub>2</sub>O was judged to yield an approximately flat baseline at 1900-1400 cm<sup>-1</sup>, and subtraction of 2 H<sub>2</sub>O was adjusted to the removal of the 2 H<sub>2</sub>O bending absorption close to 1220cm<sup>-1</sup>.

**For scanning.**

1. The sample is grounded using an agate mortar and pestle to give a very fine powder.
2. The finely powder sample is then mixed with about 100mg dried KBr salt.
3. The mixture is then pressed under hydraulic press using a die to yield a transparent disc and measure about 13mm diameter and 0.3 mm in thickness.

**Nujol mull method:**

1. The sample is ground using an agate mortar and pestle to give a very fine powder.
2. A small amount is then mixed with nujol oil to give a paste and this paste is then applied between two sodium chloride plates.
3. The plates are then placed in the instrument sample holder ready for scanning.

**Liquids:**

1. Viscous liquids can be smeared in the cell and directly measured.
2. For dilute solutions, liquid cells and variable path length cells are employed.

**Applications:**

It is the preferred method of infrared spectroscopy. FT-IR is an important and more advanced technique. It is used to identify the functional group, to determine the quality and consistency of the sample material and can determine the amount of compounds present in the sample. It is an excellent tool for quantitative analysis.

In FT-IR infrared is passed from a source through a sample. This infrared is absorbed by the sample according to the chemical properties and some are transmitted. The spectrum that appears denotes the molecular absorption and transmission. It forms the molecular fingerprint of the sample. Like the finger print there is no two unique molecular structures producing the same infrared spectrum. It is recorded as the wavelength and the peaks seen in the spectrum indicates the amount of material present.

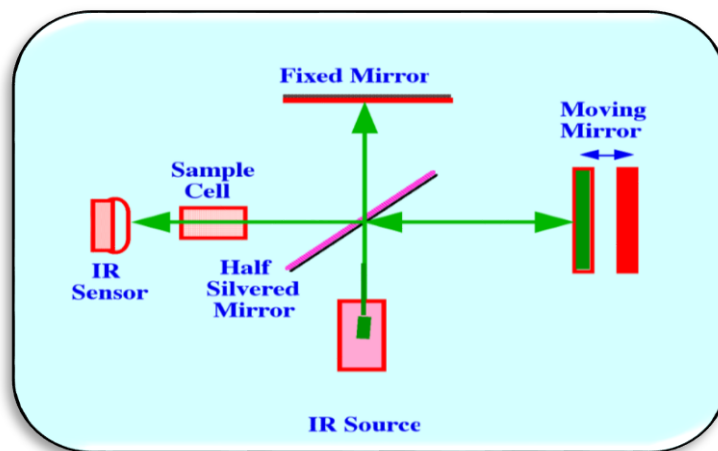
FT-IR is the most advanced and the major advantage is its

- ❖ Speed
- ❖ Sensitivity
- ❖ Mechanical Simplicity
- ❖ Internally Calibrated .

**Analytical capabilities:**

1. Identifies chemical bond functional groups by the absorption of infrared radiation which excites vibrational modes in the bond.
2. Especially capable of identifying the chemical bonds of organic materials.
3. Detects and identifies organic contaminants.
4. Identifies water, phosphates, sulphates, nitrates, nitrites, and ammonium ions.
5. Detection limits vary greatly , but are sometimes  $<10^{13}$  bonds/cm<sup>3</sup> or sometimes sub monolayer. Useful with solids, liquids, or gases.

**Fig 2.FTIR ( Fourier Transform Infrared Spectroscopy)**



**FTIR MECHANISM**

**Result:**

The result of FTIR was represented in table no -5

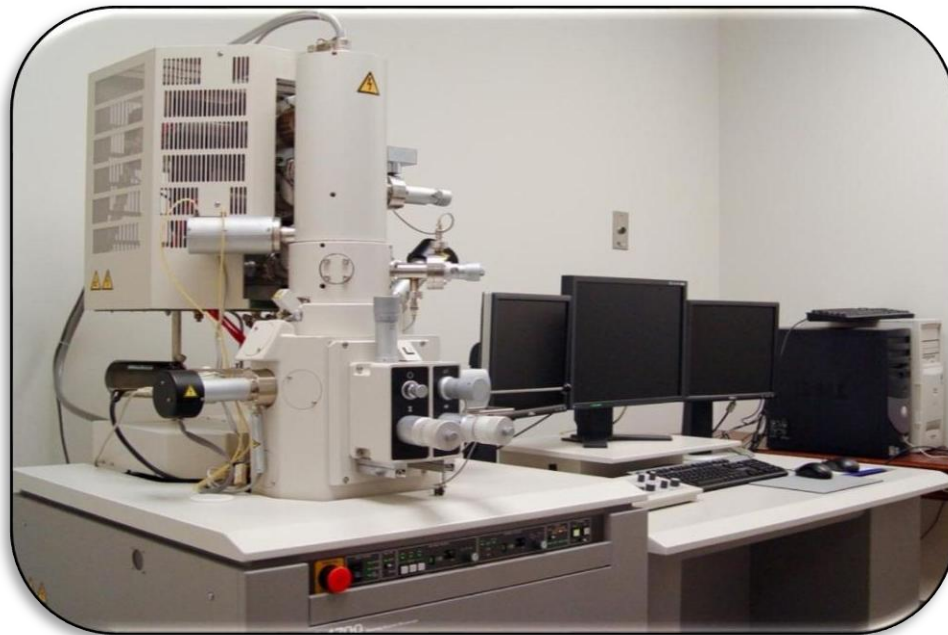
### **5.5. SEM (SCANNING ELECTRON MICROSCOPE) :**

The particle size of the Surangusa parpam was determined using high resolution scanning electron microscopy (HR SEM). The experimental procedure was done at CECRI, Karaikudi.

#### **DEFINITION**

Scanning Electron Microscopy (SEM), also known as SEM analysis or SEM microscopy, is used very effectively in microanalysis and failure analysis of solid inorganic materials. The electrons interact with atoms in the sample, producing various signals that contain information about the samples surface topography and composition. The electron beam is scanned in a raster scan pattern, and the beams position is combined with the detected signal to produce an image. It is a powerful and mature technique in the examination of materials, widely in metallurgy, geology, biology and medicine.

Scanning electron microscopy is performed at high magnifications, generates high-resolution images and precisely measures very small features and objects.



**FIG :3 SEM INSTRUMENT**

The quanta 200 FEG scanning electron microscope (SEM) is a versatile high resolution scanning electron microscope with three modes of operation namely,

1. High vacuum (HV) mode for metallic (electrically conducting) sample.
2. Low vacuum (LV) mode for insulating, ceramic, polymeric (electrically insulating)
3. Environment scanning electron microscope (ESEM) for biological samples.

Apart from giving the high resolution surface morphological images, the quanta 200 FEG also has the analytical capabilities such as detecting the presence of elements down to boron on any solid conducting materials through the energy dispersive x-ray spectrometry (EDX) providing crystalline information from the few nanometer depth of the material surface via electron back scattered detection (BSD) system attached with microscope and advanced technological PBS (WDS) for elemental analysis. EDX analysis is useful in the surface of the specimen. The EDX analysis system works as an integrated feature of a scanning electron microscope (SEM) and cannot operate on its own without the latter.

### **Principle:**

The primary electron beam interacts with the sample in a number of key ways:

- Primary electrons generate low energy secondary electrons, which tend to emphasize the topographic nature of the specimen.
- Primary electrons can be back scattered which produces images with a high degree of atomic number (Z) contrast.
- Ionized atoms can relax by electron shell to shell transitions, which lead to either X-ray emission or auger electron ejection.
- The x-ray emitted are characteristic of the elements in the top few  $\mu\text{m}$  of the sample and are measured by the EDX detector.

### **Method:**

A representative portion of each sample was sprinkled on to a double side carbon tape and mounted on aluminium stubs, in order to get a higher quality secondary electron image for SEM examination.

**Resolution :** 1.2 nm gold particle separation on a carbon substrate

**Magnification:** From a min of 12X to greater than 1,00,000 X.

**Application** : To evaluate grain size, particle size distributions , material homogeneity and inter metallic distributions.

Sample required :

- Any dimension (height or diameter) less than 10mm.
- The ideal shape of a sample was that of a button on a shirt. However, the other sizes can also be accommodated only after the discussion with the system operator.
- If the sample was not electrically conducting, it will require silver or gold coating.
- If the sample was a powder, make a normal button size pellet of the sample.
- If the sample was insulator (or) polymeric (or) electrically non conducting it needs to be coated with carbon.

**Sample preparation:**

- Sample preparation can be minimal or elaborate for SEM analysis, depending on the nature of the samples and the data required.
- Minimal preparation includes acquisition of a sample that will fit into the SEM chamber and some accommodation to prevent gaseous build-up on electrically insulating samples.
- Most electrically insulating samples are coated with a thin layer of conducting material, commonly carbon, gold or some other metal or alloy. The choice of material for conductive coatings depends on the data to be acquired.
- Carbon is most desirable if elemental analysis is a priority, while metal coatings are most effective for high resolution electron imaging applications.

**Calculation of the particle size:**

The horizontal line in the right corner of the micrograph corresponds to micro in length would be given. A comparison could be made between the length of the particles visible in the micrograph with this line and the length of the particles was calculated.

**Procedure:**

An electron beam passing through an evacuated column is focused by electromagnetic lenses onto the specimen surface. Since an electron is a charged particle. It has strong interaction with the specimen (due to coulomb interaction). So when an electron beam images on a specimen, it is scattered by atomic layers near the

surface of the specimen. As a result, the direction of electron motion changes and its energy is partially lost. Once an incident electron (primary electron) enters a substance, its direction of motion is influenced by various obstructions (multiple scattering), and follows a complicated trajectory which is far from a straight line. Also, when electrons with the same energy are incident on the specimen surface, a portion of electrons is reflected in the opposite direction (back scattered) and the remainder is absorbed by the specimen (exciting X-rays or other quanta in the process). If the specimen is sufficiently thin, the electron can pass all the way through the specimen (transmitted electrons, scattered or non scattered).

The depth at which various signals are generated due to electron beam-specimen interaction indicates the diffusion area of the signals in the specimen in addition to the local chemistry of the specimen. Secondary electrons mainly indicate information about the surface of a specimen. Since secondary electrons do not diffuse much inside the specimen, they are most suitable for observing the fine structures of the specimen surface. That is to say, sharp scanning images with high resolution can be expected from secondary electrons, because of the smaller influence on resolution by their diffusion.

As the incident electron energy increases, the probability of incident electrons colliding with elemental components of the specimen and releasing secondary electrons also increases. In other words, as the incident energy increases, the emission of electrons from the specimen also increases. However, as the energy increases beyond a certain level, the incident electrons penetrate deeper into the specimen to reach the specimen with the result that the specimen derived electrons use up most of their energy to reach the specimen surface. Consequently, the electron emission yield decreases. Therefore, the peak secondary electron emission yield occurs at a specific energy level of the incident electrons.

In order to verify the existence of a substance and recognize its shape, the image contrast must be well defined. In other words, even if a system boasts extremely high resolution, if image contrast is poor, it would be extremely difficult to determine the existence of a substance, let alone recognize its shape, another important feature of the SEM is the three dimensional appearance of the specimen image, which is a direct result of the large depth of field.

**Advantages of SEM:**

1. It gives detailed 3D and topographical imaging and the versatile information garnered from different detectors.
2. This instrument works very fast.
3. Modern SEMs allow for the generation of data in digital form.
4. Most SEM samples require minimal preparation actions.

**Disadvantages of SEM:**

1. SEMs are expensive and large.
2. Special training is required to operate an SEM.
3. The preparation of samples can result in artifacts.
4. SEMs are limited to solid samples.
5. SEMs carry a small risk of radiation exposure associated with the electrons that scatter from beneath the sample surface.

**SEM ANALYSIS APPLICATIONS**

The signals generated during SEM analysis produce a two-dimensional image and reveal information about the sample including:

- ❖ External morphology (texture)
- ❖ Chemical composition (when used with EDS)
- ❖ Orientation of materials making up the sample
- ❖ The EDS component of the system is applied in conjunction with SEM analysis
- ❖ Determine elements in or on the surface of the sample for qualitative information
- ❖ Measure elemental composition for semi-quantitative results
- ❖ Identify foreign substances that are not organic in nature and coatings on metal
- ❖ SEM Analysis with EDS – qualitative and semi-quantitative results
- ❖ Magnification – from 5x to 300,000x
- ❖ Sample Size – up to 200 mm (7.87 in.) in diameter and 80 mm (3.14 in.) in height  
Materials analysed – solid inorganic materials including metals and minerals.



## **THE SEM ANALYSIS PROCESS**

Scanning Electron Microscopy uses a focused beam of high-energy electrons to generate a variety of signals at the surface of solid specimens. In most SEM microscopy applications, data is collected over a selected area of the surface of the sample and a two-dimensional image is generated that displays spatial variations in properties including chemical characterization, texture and orientation of materials. The SEM is also capable of performing analyses of selected point locations on the sample. This approach is especially useful in qualitatively or semi-quantitatively determining chemical compositions, crystalline structure and crystal orientations.

The EDS detector separates the characteristic X-rays of different elements into an energy spectrum and EDS system software is used to analyse the energy spectrum in order to determine the abundance of specific elements. A typical EDS spectrum is portrayed as a plot of X-ray counts vs. energy (in keV). Energy peaks correspond to the various elements in the sample. Energy Dispersive X-ray Spectroscopy can be used to find the chemical composition of materials down to a spot size of a few microns and to create element composition maps over a much broader raster area. Together, these capabilities provide fundamental compositional information for a wide variety of materials, including polymers. In scanning electron microscope high energy electron beam is focused through a probe towards the sample material. Variety of signals was produced on interaction with the surface of the sample. This results in the emission of electrons or photons and it is collected by an appropriate detector.

## **SEM MECHANISM**

The types of signal produced by a scanning electron microscope include

- ❖ Secondary electrons
- ❖ back scattered electrons
- ❖ characteristic x-rays, light
- ❖ specimen current
- ❖ Transmitted electrons.

This gives the information about the sample and it includes external morphology, texture, its crystalline structure, chemical composition and it displays the shape of the sample.

**EDAX: (Energy Dispersive X-Ray Analysis)**

Energy Dispersive X-Ray Analysis is also known as EDS or EDAX. It is an X-Ray technique used to detect the composition of elements present in the given material. It has its attachments to electron microscopy instruments like scanning electron microscopy (SEM) or transmission electron microscopy (TEM) as the imaging competence of the microscope identifies the sample material.



**Figure : 4 EDAX instrument**

The data produced by the EDAX analysis consists of the spectra containing the elements present in the given sample which is being analysed. It is also possible to get the elemental mapping and image analysis of the sample.

EDAX technique is a non-destructive and can be qualitative, quantitative and provide spatial distribution of the elements.

**Results:**

The results were represented in table no.6

## 5.6. ULTRAVIOLET – VISIBLE SPECTROSCOPY:-

UV spectroscopy is an important tool in analytical chemistry. The other name of UV (Ultra violet) spectroscopy is Electronic spectroscopy as it involves the promotion of the electrons from the ground state to the higher energy or excited state.

### Introduction to UV spectroscopy:-

UV spectroscopy is type of absorption spectroscopy in which light of ultra violet region (200-400nm) is absorbed by the molecule. Absorption of the ultra violet radiations results in the excitation of the electrons from the ground state to higher energy state.

### Principle of UV spectroscopy:-

UV spectroscopy obeys the Beer-Lambert law, which states that: when a beam of monochromic light is passed through a solution of an absorbing substance, the rate of decrease of intensity of radiation with thickness of the absorbing solution is proportional to the incident radiation as well as the concentration of the solution.

### Procedure:-

Monochromators generally composed of prisms and slits. The most of the spectrometers are double beam spectrophotometers. The radiation emitted from the primary source is dispersed with the help of rotating prisms. The various wave lengths of the light source which are separated by the prism are then selected by the slits such the rotation of the prism results in a series of continuously increasing wave length to pass through the slits for recording purpose. The beam selected by the slit is monochromatic and further divided into two beams with the help of another prisms.



**Figure: 5 UV instrument**

**Uses:****Identification of an unknown compound**

An unknown compound can be identified with the help of UV spectroscopy. The spectrum of unknown compound is compared with the spectrum of a reference compound and if both the spectrums coincide then it confirms the identification of unknown substance.

**Determination of the purity of a substance:**

Purity of a substance can also be determined with the help of UV spectroscopy. The absorption of the sample solution is compared with the absorption of the reference solution. The intensity of the absorption can be used for the relative calculation of the purity of sample solution.

## 6. PHARMACOLOGICAL ACTIVITY

### 6.1.ANTI-INFLAMMATORY ACTIVITY OF *SURANGUSA PARPAM*

#### Aim:

To study the Anti-inflammatory effect of *Surangusa parpam* in Wistar albino rats by Carrageenan-induced rat paw edema.

#### Materials and methods:

<b>Test Substance</b>	:	<i>Surangusa parpam</i>
<b>Animal Source</b>	:	TANUVAS, Madhavaram, Chennai.
<b>Animals</b>	:	Wistar Albino Rats (Male -12, Female -12)
<b>Age</b>	:	6-8 weeks
<b>Body Weight</b>	:	140-160gm.
<b>Acclimatization</b>	:	14 days prior to dosing.
<b>Veterinary examination</b>	:	Prior and at the end of the acclimatization period.
<b>Identification of animals</b>	:	By cage number, animal number and individual marking by using Picric acid.
<b>Diet</b>	:	Pellet feed
<b>Water</b>	:	Aqua guard portable water in polypropylene bottles.
<b>Housing &amp; Environment</b>	:	The animals were housed in Polypropylene cages provided with bedding of husk.
<b>Housing temperature</b>	:	24-28°C
<b>Relative humidity</b>	:	between 30% and 70%,
<b>Air changes</b>	:	10 to 15 per hour
<b>Dark and light cycle</b>	:	12:12 hours.

#### Selection of animals:

Healthy Wistar albino rats (140- 160g) of both sexes were used for this study with the approval of the Institutional Animal Ethics Committee and obtained from the animal laboratory. IAEC approved no: NIS/IAEC-V/09082017/07

The animals kept in plastic cages and maintained at 24-28°C. All the rats were housed individually with free access to food, water and libitum. They were feed with

standard diet and kept in well ventilated animal house they also maintained with alternative dark-light cycle of 12hrs throughout the studies. Rats were allowed an acclimatization period of 14 days before actual experiments.

The rats were closely observed for any infection and if they show signs of infection they were excluded from the study. The animal experiment was performed with accordance legislation on welfare.

### **The experimental protocol**

Both sex of Adult wistar Albino rats weighing (140-160g) were used in this study. Rats were divided in to 4 groups, consisting six animals for each group.

Group I	-	Honey(10ml/kg)
Group II	-	Indomethacin (10mg/kg)
Group III	-	<i>Surangusa parpam</i> (15mg/kg)
Group IV	-	<i>Surangusa parpam</i> (40mg/kg)

Acute inflammation was induced by carrageenan. Carrageenan was administrated by sub- planter injection of 0.1 ml freshly prepared 1% suspension in right hind paw in rats. The paw volume was measured initially and then 1,2,3,4 hr after the carrageenan injection by using plethysmoGraphic method.

All the results were reported as mean + SEM. They were further analyzed using Two way analysis of variables (ANOVA) followed by Tukey's multiple comparison test.

**Result :** The result of anti Inflammatory activity were showed in table – 7

## **6.2.ANTI-HISTAMINE ACTIVITY OF *SURANGUSA PARPAM*:**

### **Aim :**

To evaluate the anti- histamine activity of Surangusa parpam in Wistar albino rats by Evans dye method

### **Materials and methods :**

Test Substance	:	Surangusa parpam
Animal Source	:	The Tamilnadu Veterinary and Animal sciences university, madhavaram.
Age	:	8-12weeks
Body Weight	:	250-250gm
Acclimatization	:	14 days prior to dosing
Veterinary examination	:	Prior and at the end of the acclimatization period
Identification of animals	:	By cage number, animal number and individual marking by using Picric acid.
Diet	:	Pellet feed
Water	:	Agua guard portable water in polypropylene bottles
Housing & Environment	:	The animals were housed in Polypropylene cages provided with bedding of husk.
Housing temperature	:	24-28 <sup>0</sup> C
Relative humidity	:	Between 30% AND 70%
Air changes	:	10 To 15 hours
Dark and light cycle	:	12:12 hours

### **Selection of animals:**

Healthy wistar albino rats (150-200 gm) of both sex were used for this study with the approval of the institutional animal ethics committee and obtained from the animal laboratory IAEC approved no IAEC/LI/24/CLBMCP/2017

The animals were kept in plastic cages and maintained at 24-28 degree C. all the rats were housed individually with free access to food, water and libitum. They were feed with standard diet and kept in well ventilated animal house they also

maintained with alternative dark light cycle of 12 hrs throughout the studies. Rats were allowed an acclimatization period of 14 days before actual experiments.

The rats were closely observed for any infection and if they show signs of infection they were excluded from the study. The animal experiment was performed with accordance legislation on welfare.

#### **Experimental design for Evan's dye method:**

The animals were divided into 4 groups. Each group has 6 animals.

- |           |   |  |
|-----------|---|--|
| Group I   | - | Vehicle control (honey)                        |
| Group II  | - | Standard drug Cetirizine (20 mg/kg)            |
| Group III | - | Received test drug surangusa parpam (15 mg/kg) |
| Group IV  | - | Received test drug surangusa parpam (35 mg/kg) |

#### **Procedure : vascular permeability test in rats**

Immediately after an i.v injection of 1 ml of 1% evans blue in physiological saline, two sites on one side of the shaved back of animals were injected intradermally with 0.1 ml of physiological saline containing 0.1 micro gram histamine, contralateral sites were injected intradermally with an equal volume of physiological saline (the control skin areas). Surangusa parpam is given orally 30 mins before to the injection of phlogistic agents. Thirty minutes later, the animals were sacrificed by overdose of anesthesia and the skin was removed. Exudation of dye was calculated by subtracting the amount determined in the control skin area and expressed as the mean of two values obtained in each animal.

**Result :** The result of anti histamine activity were showed in table – 9



### **6.3. ANTI PYRETIC ACTIVITY OF *SURANGUSA PARPAM*:**

#### **AIM:**

To study the anti pyretic activity effect of Surangusa parpam in Wistar albino rats by Brewer's Yeast induced pyrexia.

#### **MATERIALS AND METHODS :**

Test Substance	:	Surangusa parpam
Animal Source	:	The Tamilnadu Veterinary and Animal Sciences, University, Madhavaram.
Animal	:	Wistar albino rats (male-12, female 12)
Age	:	6-8weeks
Body Weight	:	140-160gm
Acclimatization	:	14 days prior to dosing
Veterinary examination	:	Prior and at the end of the acclimatization period
Identification of animals	:	By cage number, animal number and individual marking by using Picric acid.
Diet	:	Pellet feed
Water	:	Aqua guard portable water in polypropylene bottles
Housing & Environment	:	The animals were housed in Polypropylene cages provided with bedding of husk.
Housing temperature	:	24-28 <sup>0</sup> C
Relative humidity	:	Between 30% AND 70%
Air changes	:	10 To 15 hours
Dark and light cycle	:	12:12 hours

#### **Selection of animal :**

Healthy wistar albino rats (140-160 gm) of both sex were used for this study with the approval of the institutional animal ethics committee and obtained from the animal laboratory IAEC approved no IAEC/LI/24/CLBMCP/2017

The animals were kept in plastic cages and maintained at 24-28 degree C. all the rats were housed individually with free access to food, water and libitum. They

were feed with standard diet and kept in well ventilated animal house they also maintained with alternative dark light cycle of 12 hrs throughout the studies. Rats were allowed an acclimatization period of 14 days before actual experiments.

The rats were closely observed for any infection and if they show signs of infection they were excluded from the study. The animal experiment was performed with accordance legislation on welfare.

### **The experimental protocol:**

#### **Animal grouping:**

Both sex of Adult wistar Albino rats weighing (140-160g) were used in this study. Rats were divided into 4 groups, consisting six animals for each group.

Group I	-	Control-Honey
Group II	-	Received Standard drug Paracetamol (50 mg/kg orally)
Group III	-	Received Surangusa parpam (15 mg/kg orally)
Group IV	-	Received Surangusa parpam (35 mg/kg orally)

#### **Brewer's yeast induced hyper pyrexia method:**

The animals were fasted overnight with free access to water prior to the experimental procedure. The normal temperature of each rat in four hours was measured rectally at one hour interval on a clinical thermometer.

Before yeast injection the based rectal temperature of rats was recorded by inserting the clinical thermometer to a depth of 2 cm into the rectum and after recording animals were given subcutaneous injection of 10 ml/kg of 20% w/v yeast suspended in normal saline below the nape of the neck for elevation of body temperature of rats.

After 8 hours of yeast injection, rats which shows a rise in temperature of atleast 1<sup>0</sup>C Were taken for the study. The honey was administered orally to the control groups of animals and paracetamol at the dose of 150 mg/ ml was administered orally to the standard group of animals.

Surangusa parpam was administered orally at a dose of 15 mg/kg and 35 mg/kg weight to Group -III and Group IV respectively. Rectal temperature was recorded by clinical thermometer at 0,1,2,3 hrs after drug administration and tabulated.

**Evaluation of parameters:**

Anti pyretic activity was evaluated by comparing initial rectal temperature (°C) before yeast injection, with rectal temperature (°C) after 8 hours of yeast injection at different time intervals

**Statistical analysis:**

All the result were reported as mean  $\pm$ SD. They were further analyzed using one way analysis of variable (ANOVA) followed by Dunnet's multiple comparison test.

**Result :** The result of anti Pyretic activity were showed in table – 8

## 7.RESULTS:

Many studies have been carried out to bring the efficacy and potency of the drug *SURANGUSA PARPAM*. The study includes literary collections, organoleptic character, physicochemical analysis, FTIR, UV, SEM-EDAX, and pharmacological study. The drug *SURANGUSA PARPAM* has been selected from the text “*ANUBOGA VAITHIYA*

*NAVANEETHAM PART III- PG.NO -90 ”.*

- ❖ Botanical aspect explains the active principle and medicinal uses of the plants.
- ❖ Gunapadam review brings the effectiveness of the drug in the management of respiratory disorder.
- ❖ The pharmacological review explains about the evaluation Of Anti inflammatory, Anti histamine and Anti pyretic Activities.

### **Standardization of the test drug**

Traditional remedies is advantageous, it does suffer some limitations. The main limitation is the lack of standardization of raw materials, of processing methods and of the final products, dosage formulation, and the non- existence of criteria for quality control. Standardization of the drug is more essential to derive the efficacy, potency of the drug by analyzing it through various studies. Following tables and charts are the results of physicochemical and chemical analysis. Physical characterization and estimation of basic and acidic radicals have been done and tabulated. pharmacological activity of the drug were derived. Its result has been tabulated below.

## ANALYTICAL STUDY OF SURANGUSA PAMPAM

### 1. ORGANOLEPTIC CHARACTER

**Table: 1. Organoleptic characters of Surangusa pampam**

<b>Colour</b>	White
<b>Odour</b>	Pungent
<b>Taste</b>	Characteristic taste
<b>Texture</b>	Powder

### 2. PHYSICO-CHEMICAL ANALYSIS

**Table: 2. Physico-chemical properties of surangusa pampam**

S.No.	Parameters	Results
1	<b>LOD</b>	1%
	<b>Ash value</b>	
2	<b>a. Total ash (w/w)</b>	98.52%
	<b>b. Acid insoluble ash (w/w)</b>	23.64%
	<b>c. Water Soluble ash (w/w)</b>	5.42%
3	<b>Extractive values</b>	
	<b>a. Alcohol successive soluble (w/v)</b>	15.47%
4	<b>PH</b>	7.5

#### **Interpretation Ash:**

Ash constitutes the inorganic residues obtained after complete combustion of a drug. Thus Ash value is a validity parameter describe and to assess the degree of purity of a given drug

#### **Total ash:**

Total ash value of plant material indicated the amount of minerals and earthy materials present in the plant material. The total inorganic content (ammonium, potassium, calcium, chloride, iron, etc.,) present in the drug is measured through the Total ash value and it is of 98.52% for Surangusa pampam

**Acid insoluble ash:**

The acid insoluble ash value of the drug denotes the amount of siliceous matter present in the plant. The quality of the drug is better if the acid insoluble value is low. It is 23.64% for Surangusa parpam

**Water soluble ash:**

Water-soluble ash is the part of the total ash content, which is soluble in water. It is 5.42% for Surangusa parpam

**EXTRACTIVE VALUES**

- ❖ These are indicating the approximate measure of chemical constituents of crude drug.
- ❖ The percentage of soluble matters present in the drug is determined by the values of water extractive and ethanol extractive.
- ❖ Based on the extractive value suitable solvent can be selected. It also gives the percentage of drug which will correlate with the metabolism reactions.
- ❖ Water-soluble extractive value plays an important role in evaluation of crude drugs
- ❖ The alcohol-soluble extractive value was also indicative for the same purpose as the water- soluble extractive value

**Loss on drying**

- ❖ The total of volatile content and moisture present in the drug was established in loss on drying.
- ❖ Moisture content of the drug reveals the stability and its shelf-life.
- ❖ High moisture content can adversely affect the active ingredient of the drug.
- ❖ Thus low moisture content could get maximum stability and better shelf life.

**pH:**

- ❖ It is a measure of hydrogen ion concentration; it is the measure of the acidic or alkaline nature. 7.0 is neutral, above 7.0 is alkaline and below is acidic.  
The pH of the drug Surangusa parpam is 7.5 which is slightly alkaline in nature and it is essential for its bioavailability and effectiveness

### 3. CHEMICAL ANALYSIS

The Chemical analysis shows the presence of Phosphate, sulphate, chloride, carbonate sulphide, Iron, Zinc, Calcium, fluoride, oxalate, ammonium, copper, aluminium, Magnesium, Potassium, Alkaloids and tannic acid in *Surangusa parpam*.

**Table: 3. Chemical Analysis of *Surangusa parpam*-Acid Radicals**

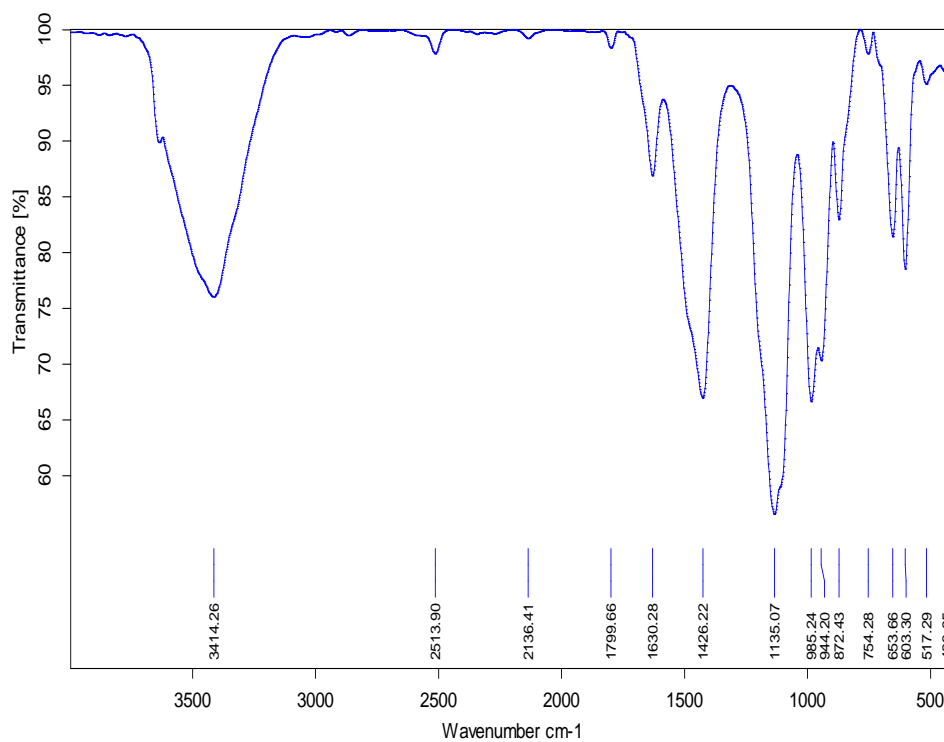
S.NO	Parameters	Results
1.	Silicate	Absent
2.	Sulphate	<b>Present</b>
3.	Chloride	<b>Present</b>
4.	Phosphate	<b>Present</b>
5.	Carbonate	<b>Present</b>
6.	Nitrate	Absent
7.	Sulphide	<b>Present</b>
8.	Oxalate	<b>Present</b>
9.	Nitrite	Absent
10.	Borate	Absent
11.	Lead	Absent
12.	Copper	<b>Present</b>
13.	Aluminium	<b>Present</b>

#### **Interpretation**

The acidic radicals test shows the presence of **Phosphate, chloride, sulphate, carbonate, copper, aluminium, oxalate and sulphide.**

#### 4. FT-IR (Fourier Transform Infra-Red) Spectroscopy

3414, 2513, 2136, 1799, 1630, 1426, 1136, 986, 944, 872, 754, 663, 603, 517, 433



E:\EXTERNAL\Chennai\SAMPLE-1 01-04-2019.0

SAMPLE-1 01-04-2019

Instrument type and / or accessory

13/11/2007

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**Figure 6. FT-IR Spectrum of Surangusa parpam**



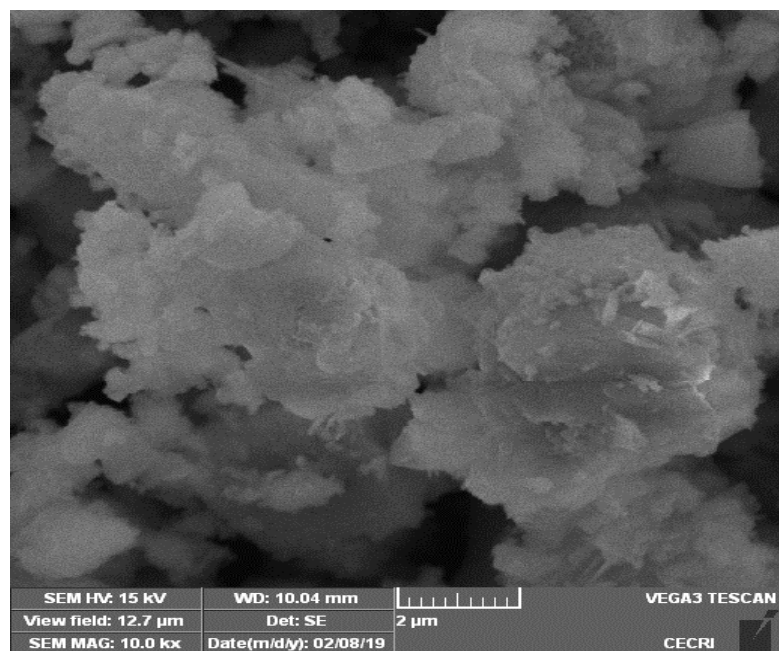
## Interpretation

**Table – 5 VIBRATIONAL MODES AND FUNCTIONAL GROUP OF SURANGUSA PARPAM IN FTIR :**

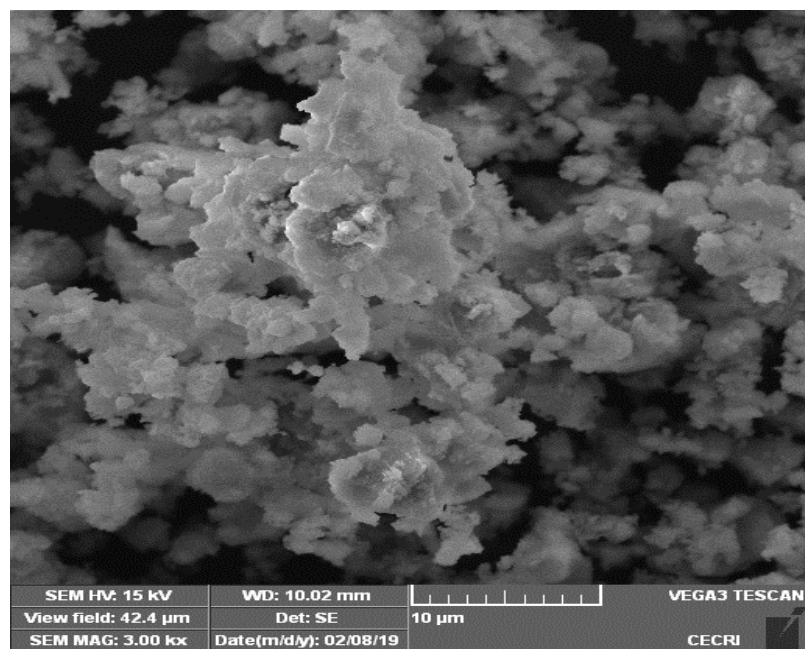
Wave number (cm-1)	Vibrational modes of <i>surangusa Parpam</i> in IR region	Functional group
<b>3414</b>	O–H stretch, H–bonded	alcohols, phenols
<b>2513</b>	O–H stretch	carboxylic acids
<b>2136</b>	–C≡C– stretch	Alkynes
<b>1630</b>	N–H bend	1° amines
<b>1426</b>	C–C stretch	Aromatics
<b>1136</b>	C–N stretch	aliphatic amines
<b>986</b>	=C–H bend	Alkenes
<b>944</b>	O–H bend , =C–H bend	carboxylic acids, alkenes
<b>872</b>	N–H wag , C–H “oop”	1°, 2° amines, aromatics
<b>754</b>	C–Cl stretch , N–H wag , C–H “oop”	alkyl halides, 1°, 2° amines, aromatics
<b>663</b>	C–Cl stretch , –C ≡C–H:C–H bend	alkyl halides, alkynes
<b>603</b>	C–Cl stretch	alkyl halides
<b>517</b>	C–Br stretch	alkyl halides

In the FT-IR Spectra analysis, this Surangusa Parpam sample exhibits the peak value shows in Table 1 at the wave number 3414, 2513, 2136, 1630, 1426, 1136, 986, 944, 872, 754, 663, 603, 517 having O-H Stretch , –C≡C– stretch , N–H bend , C–C stretch , C–N stretch , =C-H Stretch, O–H stretch , N–H wag , C–H “oop” , C–Cl stretch , C-Br Stretch. This indicates the presence of some organic functional groups such as alcohols, phenols, amines, alkynes, carboxyl groups, alkyl halides, alkenes.

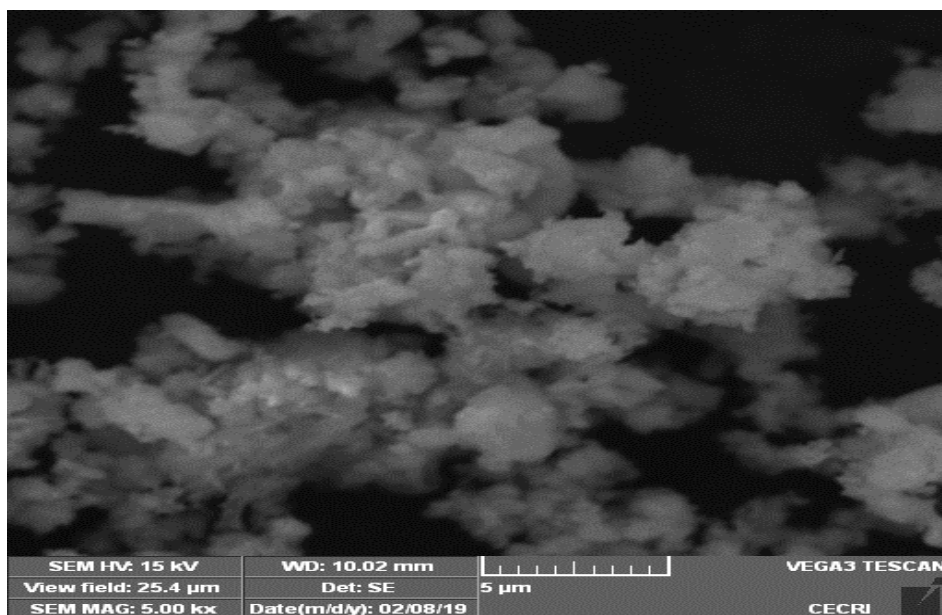
## 5. SEM WITH EDAX:



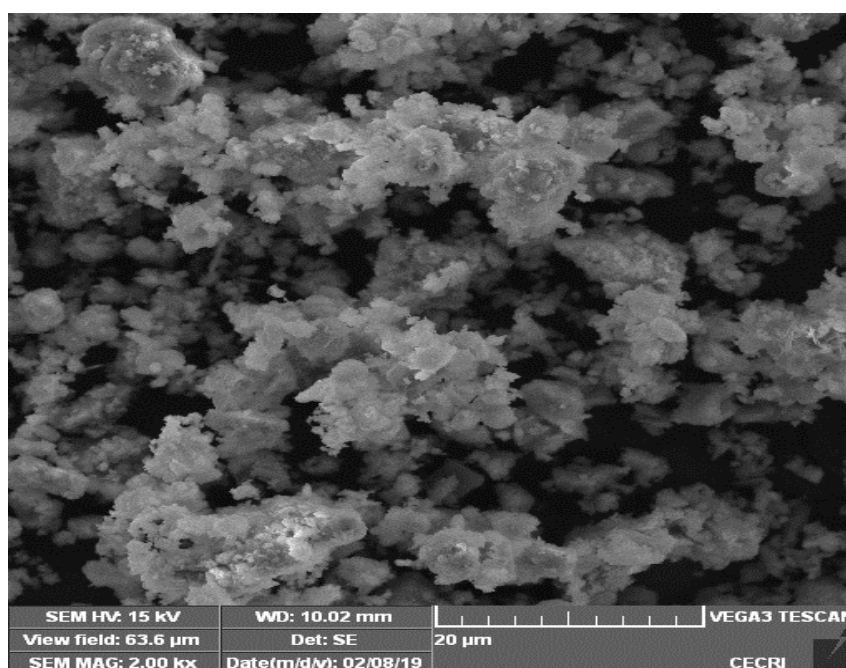
**Fig 7: Showing SEM report of SP (10KX magnification)**



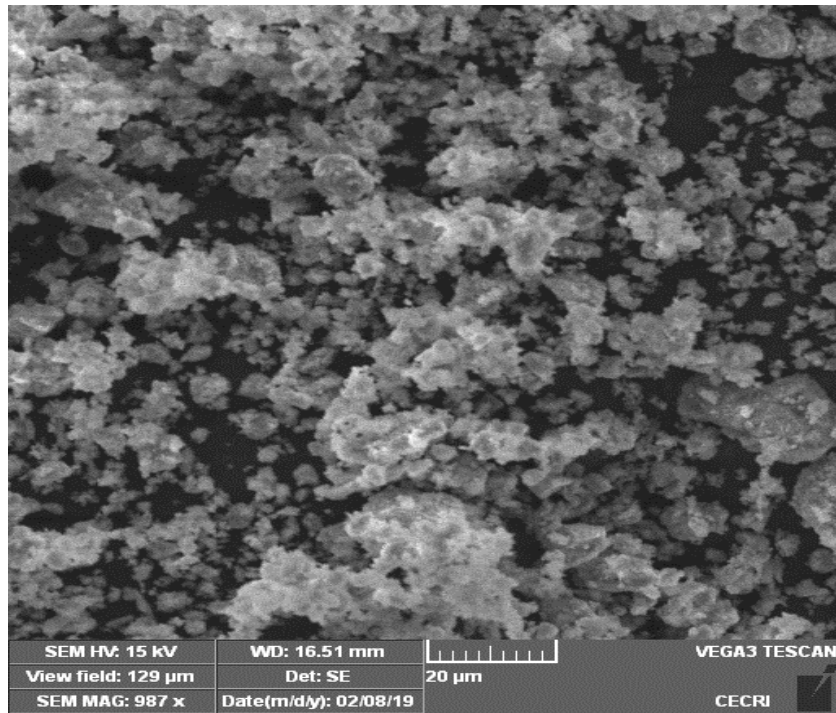
**Fig 8: Showing SEM report of SP (3KX magnification)**



**Fig 9 : Showing SEM report of SP (5 KX magnification)**



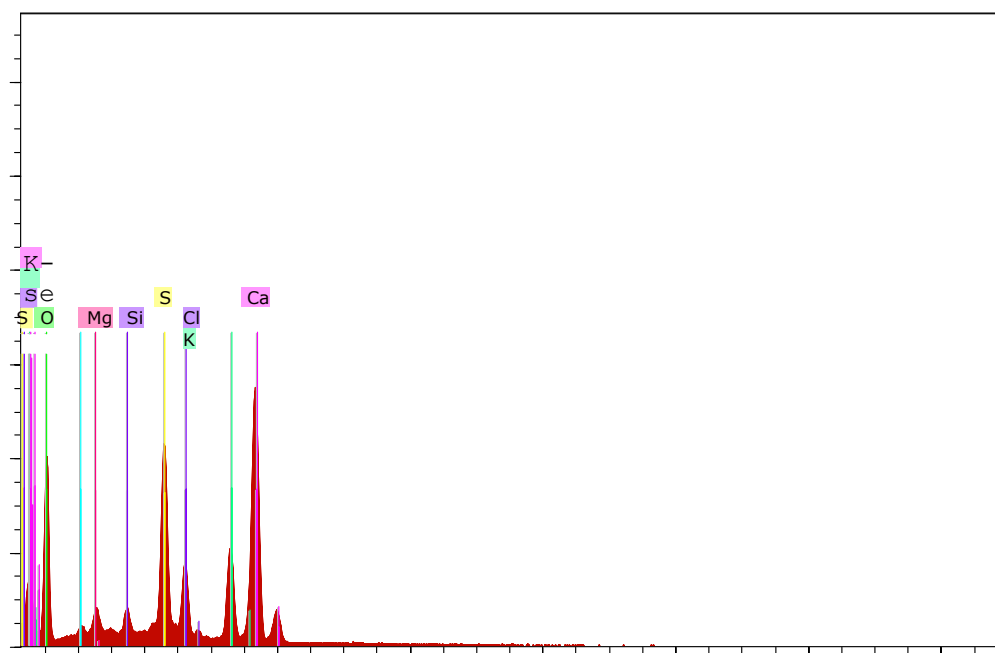
**Fig 10 : Showing SEM report of SP (2KX magnification)**



**Fig 11: Showing SEM report of SP (987X magnification)**

## EDAX :

### Elemental analysis of Surangusa parpam:



**Table:6**

Spectrum: Acquisition 8403

El	AN	Series	unn. C	norm. C	Atom. C	Error (1 Sigma)	K fact.	Z corr.	A corr.	F corr.
			[wt.%]	[wt.%]	[at.%]		[wt.%]			
O	8	K-series	33.76	46.36	65.07		4.39	0.651	0.712	1.000
Ca	20	K-series	19.78	27.16	15.22		0.62	0.137	1.967	1.000
S	16	K-series	7.01	9.62	6.74		0.28	0.049	1.924	1.000
K	19	K-series	5.84	8.02	4.60		0.21	0.036	2.104	1.000
Cl	17	K-series	3.24	4.45	2.82		0.14	0.022	1.949	1.000
C	6	K-series	1.22	1.67	3.12		0.36	0.039	0.428	1.000
Mg	12	K-series	0.87	1.20	1.11		0.08	0.009	1.363	1.000
Si	14	K-series	0.68	0.94	0.75		0.06	0.006	1.655	1.000
Na	11	K-series	0.43	0.59	0.57		0.06	0.005	1.173	1.000

## **INTERPRETATION**

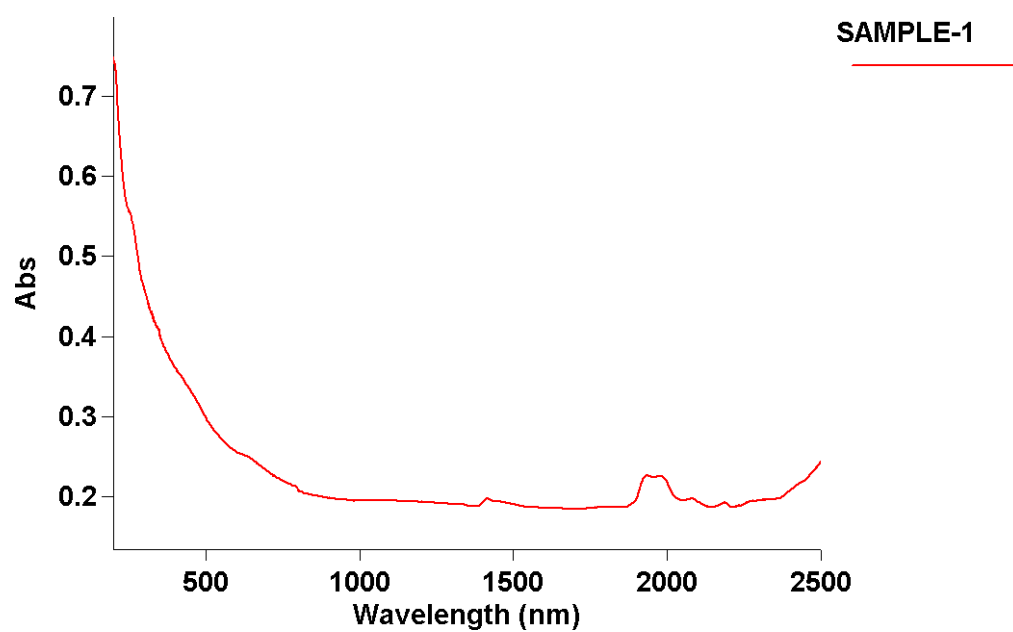
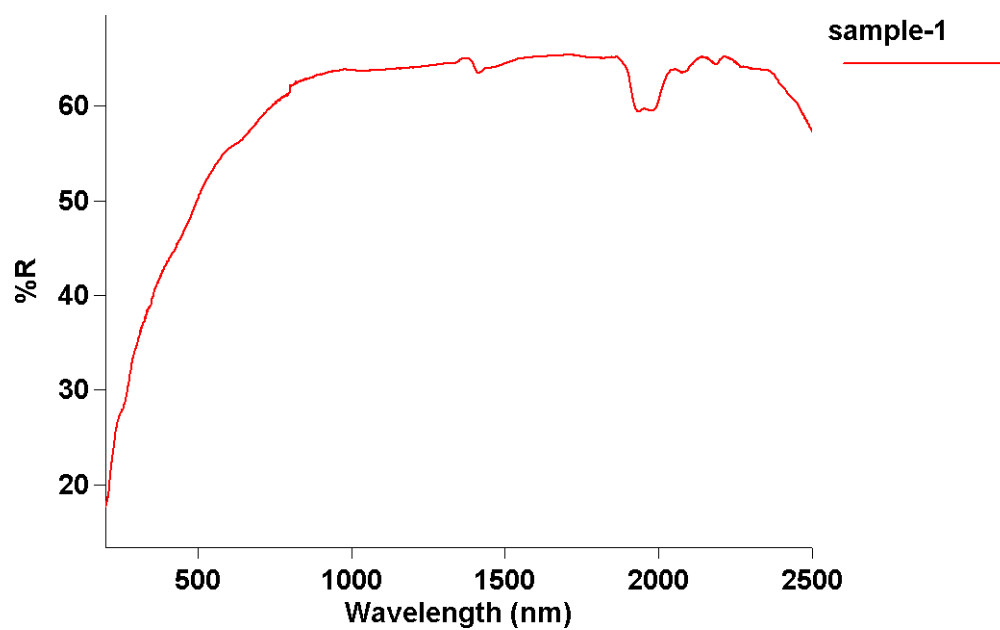
### **SEM**

The SEM photographs shows that the size of the particle is in nanometers which will promotes the easy or quick assimilation of the drug and thereby improving the efficacy.

### **EDAX**

Energy dispersive x-ray analysis (EDAX) of KKK was carried out and the elements present like Oxygen, chloride, potassium, Sulphur and Calcium were estimated. From the spectra atom percentage of the elements are found to be as follows. Oxygen= 46.36%, calcium=27.16%, chloride=4.45%, potassium=8.02%, sulphur=9.62%. SEM and EDAX provide good estimate of the concentration of main elements in the drug. Furthermore, it provides useful information in the distribution of the elements forming the drug and their sample chemical form.

## 6. UV ULTRAVIOLET – VISIBLE SPECTROSCOPY:-

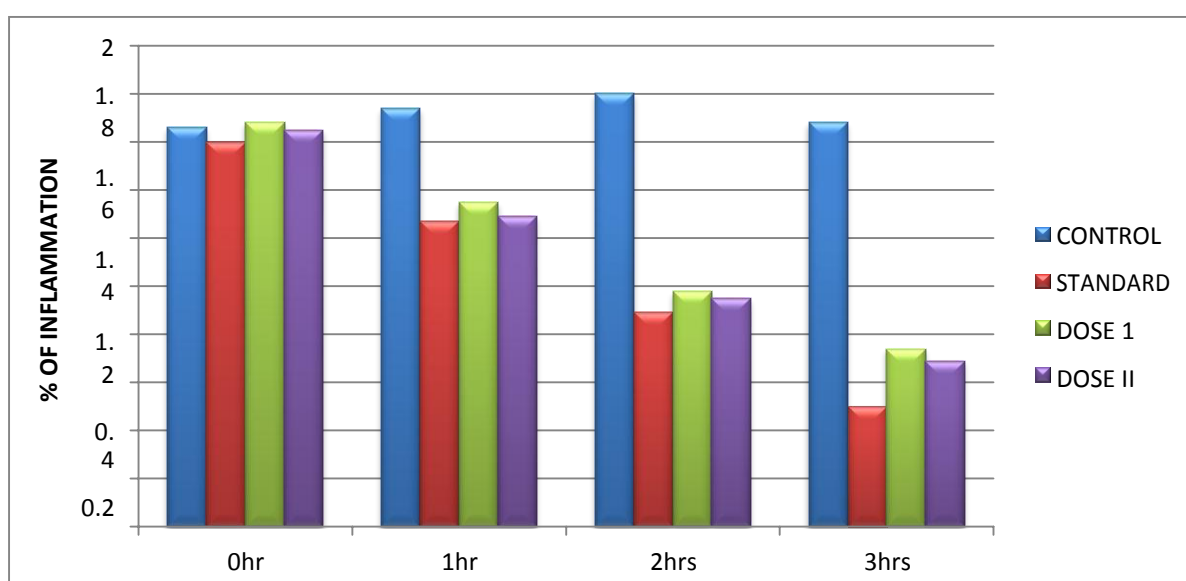


**7. Anti inflammatory activity of Surangusa parpam by carrageenan induced paw oedema in wister albino rats.**

**Table 7:**

Treatment	Percentage of inflammation after carrageenan injection at different hr			
	0hr	1hr	2hrs	3hrs
Control(honey)	1.66±1.09	1.74±0.73	1.80±0.71	1.68±0.05
Indomethacin 10mg/kg	1.60±0.82	1.27±0.87	0.89±0.81 <sup>*</sup>	0.50±0.72 <sup>**</sup>
<b>Surangusa parpam</b> 15mg/kg	1.68±0.41	1.35±0.13	0.98±0.69	0.74±0.24 <sup>**</sup>
<b>Surangusa parpam</b> 35mg/kg	1.65±0.10	1.29±0.95	0.95±0.63 <sup>*</sup>	0.69±0.56 <sup>**</sup>

Values are Mean ± SEM; n = 6 animals in each group: <sup>\*</sup> P<0.05, <sup>\*\*</sup> P< 0.01, <sup>\*\*\*</sup> P<0.001 is considered significant when compared with control rats and followed by one way ANOVA.





**Result of anti inflammatory activity:**

Surangusa parpam at 15 mg/kg dose showed significant anti inflammatory activity ( $p<0.01$ ) at 3<sup>rd</sup> hour when compared to control group. At 35mg/kg the drug showed significant ( $p<0.01$ ) at 3<sup>rd</sup> hour. Among the two doses of surangusa parpam, 35mg/kg have shown significant anti inflammatory activity ( $p<0.01$ ) when compared with control group.

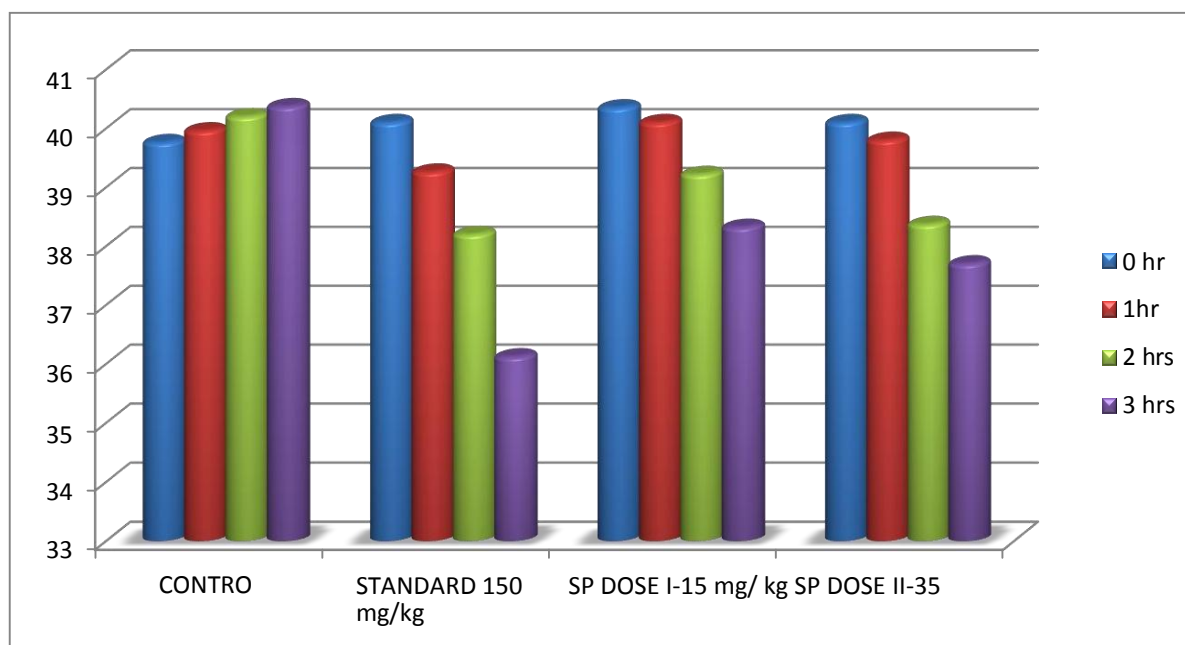
**Conclusion:**

Thus it was concluded that administration of **Surangusa parpam** the dose of 35mg/kg/ showed significant Anti inflammatory ( $p<0.01$ ) activity when compare to the control group.

**8. Anti pyretic activity of Surangusa parpam by Brewer's yeast induced method in wistar albino rats.**

**Table 8:**

Groups	Initial rectal temp <sup>0</sup> C	Rectal temp in <sup>0</sup> C after 8 hrs of yeast injection			
		0 hr	1 hr	2 hrs	3 hrs
<b>Group I</b> <b>Control</b> <b>Honey (p.o)</b>	36.97±0.2 7	39.76±0.2 4	39.96±0.22	40.20±0.10	40.37±0.15
<b>Group II</b> <b>Standard</b> <b>Paracetamo</b> <b>I</b> <b>(150 mg/ kg)</b> <b>(p.o)</b>	35.50±0.2 1	40.10±0.2 2	39.26±0.23 *	38.20±0.21 *	36.12±0.21 **
<b>Group III</b> <b>SP dose-I</b> <b>(15mg/kg)</b> <b>(p.o)</b>	36.80±0.2 7	40.35±0.3 1	40.10±0.32	39.22±0.45 *	38.32±0.21 **
<b>Group IV</b> <b>SP Dose-II</b> <b>35mg/kg)</b> <b>(p.o)</b>	36.72±0.4 0	40.10±0.3 2	39.80±0.40 *	38.37±0.38 *	37.70±0.28 **



Values are Mean  $\pm$  SEM; n = 6 animals in each group: \*  $P < 0.05$ , \*\*  $P < 0.01$ , \*\*\*  $P < 0.001$  is considered significant when compared with control rats. The results were analyzed by one way ANOVA followed by Dunnet's test.

Anti pyretic activity of surangusa parpam were evaluated using brewer's yeast induced hyperpyrexia in rats. The subcutaneous injection of yeast suspension markedly elevated the rectal temperature at the 8<sup>th</sup> hour after administration. The result obtained from the study showed that there was significant increase in the body temperature of rats injected with brewer's yeast. The antipyretic effect started as early as the first hour after administration, and the effect was maintained for three hours after its administration.

Rats treated with the standard drug paracetamol (150mg/kg) has shown maximum reduction in rectal temperature during 3<sup>rd</sup> hour after injection of brewers yeast. It was found that surangusa parpam at doses of 15mg/kg showed significant ( $p < 0.01$ ) anti pyretic activity at 3<sup>rd</sup> hour and 35mg/kg caused better significant ( $p < 0.001$ ) lowering of body temperature when compared to the control group animals at 3<sup>rd</sup> hour. Inhibition of prostaglandin synthesis could be the possible mechanism of anti pyretic action as that of paracetamol. Also, there are several mediators or multiprocesses underlining the pathogenesis of fever. Inhibition of any of these mediators may bring about anti pyretic activity.

### **Result:**

**Surangusa parpam** the dose of 35mg/kg at 3<sup>rd</sup> hour showed better significant Anti pyretic ( $p < 0.001$ ) activity when compared with control rats.

**9. Anti histamine activity of Surangusa parpam by Evans dye method in albino Wistar rats.**

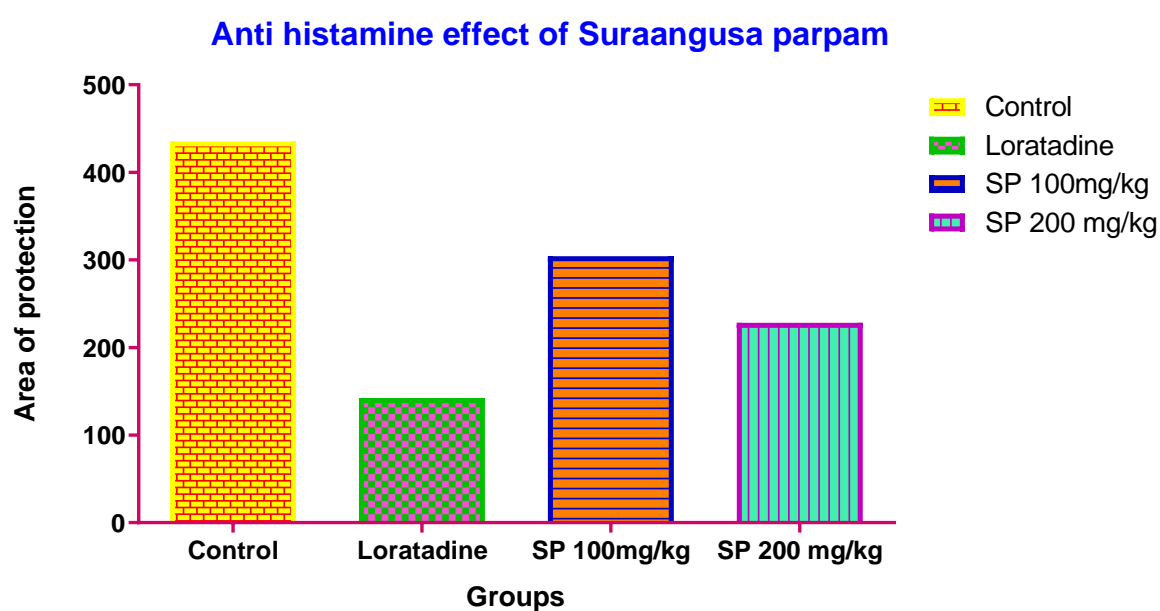
**Area of protection = control area – area of exudation of dye**

**Grouping: Wistar rats were used for the study n=6nos**

Group I	Control group
Group II	Standard drug Loratadine 20mg/kg
Group III	Suraangusa parpam 100mg/kg
Group IV	Suraangusa parpam 200mg/kg

Table 9:

S.no	Grouping	Area of protection from exudation of Dye in mm
1	Control	450.12±0.32
2	Loratadine (STD)	142.23±0.07
3	Surangusa parpam 100mg	304.24±0.12
4	Surangusa parpam 200mg	228.32±0.18



Values are Mean  $\pm$  SEM; n = 6 animals in each group: \* P<0.05, \*\* P< 0.01, \*\*\*P<0.001 is considered significant when compared with control rats and followed by one way ANOVA.

**Result :**

**Surangusa parpam** the dose of 35mg/kg/ showed significant Anti histamine(p<0.001) activity when compare to the control group.

## 8. DISCUSSION

The drug **Surangusa parpam** was selected from the Siddha literature “ANUBOGA VAITHIYA NAVANEETHAM-part III page no-90” to validate the (*Anti inflammatory, Anti histamine and Antipyretic activity*) in an animal model. The ingredients of the test drug was identified and authenticated by Siddha experts. The drug was prepared as per the procedure and subjected to various studies such as qualitative, quantitative, Standardization and pharmacological activities. Qualitative analysis includes Chemical analysis, Physicochemical properties of **Surangusa parpam**. From the above analysis we came to know the presence of active ingredients responsible for its activity.

### **Literary collections:**

Literary collections include drug review, which consist both Botanical aspect, Gunapadam aspect and pharmacological review are support this study.

### **Drug review:**

#### **Botanical aspect:**

Drug review about the ingredients of **Surangusa parpam** from various text books was done.

Botanical aspect explains the identification, description, active principle and medicinal uses of the plants. Siddha literatures related to the drug bring the evidence and importance of its utility in treating the fever and respiratory disorders.

#### **Gunapadam aspect**

- Gunapadam review brings the effectiveness of the drug in treating Respiratory disorders.
- Pepper by its formulation as chooranam directly used as an Anti pyretic Drug...

#### **Pharmaceutical aspect**

Pharmaceutical review describes about the parpam and its properties.

#### **Pharmacological aspect**

The pharmacological review explains about the methodology of Anti inflammatory, Anti histamine and Anti pyretic activity the drugs used and the analysis

of pharmacological activity through Carragenan induced paw edema method, Evans blue dye method and Brewer's yeast induced method.

They explained about the effective Anti pyretic, Anti inflammatory and Anti histamine activity of **Surangusa parpam**.

### **Physico chemical analysis**

- In physico chemical analysis, the pH of **Surangusa parpam** was found to be in the range of 7.5. The pH of the drug **Surangusa parpam** is 7.5 which is slightly alkaline in nature and it is essential for its bioavailability and effectiveness.
- The loss on drying value at 105°C of **Surangusa parpam** was found to be 1%w/w, hence the drug will not lose much of its volume on exposure to the atmospheric air at room temperature. It shows that the drug has more stability.
- Ash value 98.52 % it is the residue remaining after incineration that determines the inorganic substances present in the drug. Similarly it can also detect the nature of the material, whether it is adulterate or not. Hence, determination of the ash value provides an idea for judging the identity and purity of the drug.
- Decreased water soluble ash value (5.42%) indicates easy facilitation of diffusion and osmosis mechanisms.

### **Chemical analysis:**

Chemical analysis of the drug **Surangusa parpam** revealed the presence of Phosphate, sulphate, chloride, carbonate sulphide, Iron, Zinc, Calcium, fluoride, oxalate, ammonium, copper, aluminium, Magnesium, Potassium, Alkaloids and tannic acid in **Surangusa parpam**.

### **Instrumental analysis**

Based on the result **Surangusa parpam** is preferably non-toxic to human in its therapeutic dose. The standardization of the drug was evaluated by chemical analysis, characterization with elemental analysis, determination of particle size by FTIR and SEM-EDAX respectively.

### **FTIR**

From the results, the O-H Stretch at 3414 indicates a strong peak of potassium, the O-H Stretch at 2513 and 944 indicates a strong peak of sodium and potassium, N-

H bend at 1630 indicates a potassium, C–N stretch at 1136 indicates a sodium tetraborate,, C–Cl stretch at 663, 517 indicates a sodium . So, majorly this sample surangusa parpam contains potassium and sodium compounds. Sodium nitrate is used as an anti oxidant, it prevents the growth of bacteria and potassium helps to prevent hypokalemia. Overall observation in the sample Surangusa parpam is predominantly alkyne in nature. From that, we can conclude it may neutralize the acids easily. Also shows the presence of functional groups such as Alcohol, Amines, Alkenes, Alkyl Halide, carboxylic acid and alkyne groups. This FT-IR characterization results are creating the fingerprints to standardize this Siddha drug Surangusa parpam.

### **SEM-EDAX**

The SEM photographs shows that the size of the particle is in nanometers which will promotes the easy or quick assimilation of the drug and thereby improving the efficacy.

### **EDAX**

Energy dispersive x-ray analysis (EDAX) of KKK was carried out and the elements present like Oxygen, chloride, potassium, Sulphur and Calcium were estimated. From the spectra atom percentage of the elements are found to be as follows. Oxygen= 46.36%, calcium=27.16%, chloride=4.45%, potassium=8.02%, sulphur=9.62%. SEM and EDAX provide good estimate of the concentration of main elements in the drug. Furthermore, it provides useful information in the distribution of the elements forming the drug and their sample chemical form.

The elements such as sulphur, and oxygen, detected in the drug are commonly present in all the herbal drugs and arsenic originating from the primary metabolites.

### **Pharmacological studies:**

The pharmacological study was carried out in the animal model in Wistar albino rats. Three activities were seen in the drug of **Surangusa parpam**. The activities were

- ❖ Anti Inflammatory
- ❖ Anti pyretic
- ❖ Anti histamine



**Anti Inflammatory Activity:**

The Anti-inflammatory activity was evaluated using carrageenan-induced paw edema models in Wistar albino rats. The aqueous extract of **Surangusa parpam** has shown significant ( $P < 0.01$ ) inhibition of paw edema on 3rd hour at the doses of 15mg/kg and 35 mg/kg, respectively.

**Antipyretic Activity**

The Antipyretic activity of the surangusa parpam was carried out by Brewer's yeast induced method in wistar albino rats. The result indicates that surangusa parpam at doses of 15mg/kg and 35mg/kg at 3<sup>rd</sup> hour caused significant lowering of body temperature when compared to control group animals. Values are statistically significant at ( $p < 0.001$ ) doses of 35mg/kg. From these result it was obvious that Surangusa parpam has significant antipyretic activity.

**Anti histamine Activity**

Wistar albino rats of either sex were divided into 4 groups of 6 animals each. Group I received vehicle control (honey), group II received standard drug cetirizine (10mg/kg), group III and group IV at doses 100mg/kg and 200mg/kg respectively. From the results it was concluded that administration of surangusa parpam at the doses of 200mg/kg exhibited significant ( $p < 0.001$ ) anti histamine activity in wistar albino rats when compared with control

## 9.SUMMARY

- The test drug **Surangusa parpam** was selected from the siddha literature “Anuboga vaithiya navaneetham-part III its *Standardization and Pharmacological screening* (Anti inflammatory, Anti histamine and Anti pyretic activity) in an animal model. The dissertation started with an introduction explaining about the siddha concept and role of the test drug in treating respiratory disorders.
- The test drug was prepared properly by the given procedure. All the ingredients were identified and authenticated by the respective field experts.
- Review of literature in various categories was carried out. Siddha aspect, botanical aspect and pharmaceutical review disclosed about the drug and the disease. Pharmacological review was done to establish the methodologies.
- The drug was subjected to analysis such as, physicochemical, chemical analysis, Instrumental and pharmacological analysis which provided the key ingredients present in the drug thus it accounts the efficacy of the drug.
- Identification of functional groups was engaged by using Fourier Transform Infra Red spectroscopy [FTIR]
- The particle size and identification and quantitative analysis of chemical elements of Surangusa parpam were assessed by SEM with EDAX.
- The Instrumental analysis report reveals that the heavy metals like Arsenic, lead and Mercury are absent. Pharmacological study was done. It revealed the Anti-inflammatory, Anti pyretic and anti histamine activities of trail medicine in animal model viz., Wistar albino rats .This study suggests Surangusa parpam has remarkable medicinal value in the treatment of Fever and Respiratory disorders.
- Results and discussion gives the necessary justifications to prove the potency of the drug.
- Conclusion gives a complied form of the study and explains the synergistic effect of all the key ingredients and activities that supports the study.
- Thus the siddha formulation Surangusa parpam is validated for its safety and efficacy for treating fever and respiratory disorders it would be a great drug of choice.

## 10. CONCLUSION

From the literature evidence, Physico Chemical analysis, chemical analysis, Elemental analysis and Pharmacological studies, the author concludes that the drug Surangusa parpam is safe and it has significant effect in Anti-inflammatory, Anti pyretic and Anti histamine activities. It was concluded that the **Surangusa parpam** can be used effectively in the treatment of Fever and Respiratory disorders which is cost effective and easy to prepare.

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carrageenan induced paw edema in Rats

## 12. ANNEXURE:





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Affiliated to The Tamil Nadu Dr. M.G.R. Medical University, Chennai.  
Approved by Pharmacy Council of India, New Delhi, and  
All India Council for Technical Education, New Delhi

### CERTIFICATE

This is to certify that the project entitled “**Pharmacological study on Surangusa parpam in wistar rats**” submitted in partial fulfilment for the degree of M.D. (Siddha) was carried out at C.L. Baid Metha college of Pharmacy, Chennai-97, in the Department of Pharmacology during the academic year of 2018-2019.



*P. P. Muralidharan*  
Dr. P. Muralidharan



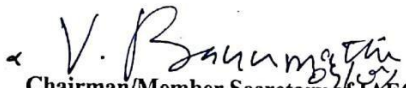
### CERTIFICATE

This is certify that the project title standardization and pharmacological screening of surangusa parpam has been approved by the IAEC. Total No. of animal sanctioned: 48 Rats  
FAEC approval NO: NIS/IAEC 7/09082017/08 (Male or Female)

Prof .Dr. V.Banumathi  
Chairman IAEC

Prof .Dr.K.Nachimuthu  
CPCSEA nominee

Signature with date

  
Chairman/Member Secretary of IAEC:

  
CPCSEA nominee:

(Kindly make sure that minutes of the meeting duly signed by all the participants are maintained by Office)

Name of the principle investigator: Dr .G.Manikandan  
Ist year pg scholar  
Department of Gunapadam  
National institute of siddha

Name of the Guide : Dr.S.Visweswaran MD (s)  
Head of the Department( i/c)  
Department of Gunapadam  
National institute of siddha

NATIONAL INSTITUTE OF SIDDHA, CHENNAI – 600047

BOTANICAL CERTIFICATE

Certified that the following plant drugs used in the Siddha formulation “*Surangusa parpam*” taken up for Post Graduation Dissertation studies by **Dr.G.Manikandan M.D.(S)**, II year, Department of Gunapadam, 2018, are identified through Visual inspection, Experience, Education & Training, Organoleptic characters, Morphology and Taxonomical methods as

*Piper nigrum* Linn. (Piperaceae). Fruit

Certificate No: NISMIB3242018



Date: 09-03-18

Authorized Signatory

**Dr. D. ARAVIND, M.D.(s), M.Sc.,**  
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---

07.02.2018

**AUTHENTICATION CERTIFICATE**

Certified that the samples submitted for identification by Dr.G.Manikandan, II year PG scholar, Dept. of Gunapadam, National Institute of Siddha, Chennai - 47, are identified as Manosilai-Arsenic disulphide , Sangu-Conch on the basis of macroscopic character.

This certificate is issued for the purpose of preparing his dissertation medicine in Gunapadam laboratory, NIS.

  
Dr. S. Visweswaran, M.D (s)  
Head of Department  
Department of Gunapadam  
National Institute of Siddha  
Tambaram Sanatorium, Chennai-47.

# ACKNOWLEDGEMENT

# INTRODUCTION

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# MATERIALS AND METHODS



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# BOTANICAL REVIEW

# SCIENTIFIC REVIEW

# ANALYTICAL STUDY OF SURANGUSA PARPAM

# PHARMACEUTICAL REVIEW

# ORGANOLEPTIC EVALUATIONS

# PHYSICO CHEMICAL ANALYSIS



# CHEMICAL ANALYSIS

# FT-IR ANALYSIS

# ULTRAVIOLET – VISIBLE SPECTROSCOPY

# SEM WITH EDAX ANALYSIS

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